APPLICATION

FOR

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TITLE: USE OF AN ARRAY OF POLYMERIC SENSORS OF VARYING THICKNESS FOR DETECTING ANALYTES IN FLUIDS

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USE OF AN ARRAY OF POLYMERIC SENSORS OF VARYING THICKNESS

FOR DETECTING ANALYTES IN FLUIDS

Cross Reference To Related Applications

This application is a continuation of 09/910,243 filed July

5   20, 2001, which is a divisional of U.S. Application No.

09/442,074, filed November 16, 1999 (now U.S. Patent No.

6,387,329) which claims the benefit under 35 U.S.C. 119(e)

of the U.S. Provisional Application No. 60/108,915, filed

on November 17, 1998 and U.S. Provisional Application No.

10 60/108,674, filed on November 16, 1998, all of which are

incorporated by reference.

Statement As To Federally-Sponsored Research

The U.S. Government has certain rights in this invention

pursuant to Grant No. DAAK60-97-K-9503 awarded by DARPA and

15 Grant No. DAAG55-97-1-0187 awarded by the U.S. Army.

Field of the Invention

This invention relates generally to sensors and sensor

systems for detecting analytes in fluids and more

20 particularly sensors having electrical properties that vary

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according to the presence and concentration of analytes, and to methods of using such sensor systems.

Background of the Invention

5   There is considerable interest in developing sensors that act as analogs of the mammalian olfactory system (Lundstrom et al. (1991) *Nature* 352:47-50; Shurmer and Gardner (1992) *Sens. Act. B* 8:1-11; Shurmer and Gardner (1993) *Sens. Actuators B* 15:32). Prior attempts to produce a broadly
10   responsive sensor array have exploited heated metal oxide thin film resistors (Gardner *et al.* (1991) *Sens. Act.* B4:117-121; Gardner *et al.* (1991) *Sens. Act. B* 6:71-75), polymer sorption layers on the surfaces of acoustic wave resonators (Grate and Abraham (1991) *Sens. Act. B* 3:85-111;
15   Grate *et al.* (1993) *Anal. Chem.* 65:1868-1881), arrays of electrochemical detectors (Stetter *et al.* (1986) *Anal. Chem.* 58:860-866; Stetter *et al.* (1990) *Sens. Act. B* 1:43-47; Stetter *et al.* (1993) *Anal. Chem. Acta* 284:1-11), conductive polymers or composites that consist of regions
20   of conductors and regions of insulating organic materials (Pearce *et al.* (1993) *Analyst* 118:371-377; Shurmer *et al.* (1991) *Sens. Act. B* 4:29-33; Doleman *et al.* (1998) *Anal. Chem.* 70:2560-2654; Lonergan *et al. Chem. Mater.* 1996, 8:2298). Arrays of metal oxide thin film resistors,

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typically based on tin oxide (SnO_2) films that have been
coated with various catalysts, yield distinct, diagnostic
responses for several vapors (Corcoran et al. (1993) *Sens.
Act. B* 15:32-37). However, due to the lack of

5   understanding of catalyst function, SnO_2 arrays do not allow
deliberate chemical control of the response of elements in
the arrays nor reproducibility of response from array to
array. Surface acoustic wave resonators are extremely
sensitive to both mass and acoustic impedance changes of

10 the coatings in array elements, but the signal transduction
mechanism involves somewhat complicated electronics,
requiring frequency measurement to 1 Hz while sustaining a
100 MHZ Rayleigh wave in the crystal.

15 Although these sensors have particular advantages there
exists a need for polymer based sensor system that shows
intra-array variation without necessarily changing the
polymer itself. Such a system would allow simultaneous
determination of kinetic and equilibrium properties of an

20 analyte. The present invention fulfills these and other
needs.

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Summary of the Invention

Systematic variation in the thickness of a non-conducting, semi-conducting, and/or conductive organic material in a

5   sensor of the invention has been performed and demonstrates that the time course of response to an analyte is different depending upon the thickness of the material film. In this way it is possible to combine rapid response times on the thinnest films in order to obtain quick information on the

10   presence of an analyte as well as its identity, while simultaneously obtaining kinetic response information that allows measurement of the permeability of the analyte through the film, yielding information on the apparent diffusion coefficient as well as other important kinetic

15   information on the properties of the analyte being detected by the sensors in the array. The elapsed time required to obtain the equilibrium constant information is therefore much shorter than would be the case if the analyte were sufficiently slow-diffusing that one had to acquire

20   measurements on one non-conductive, semi-conductive and/or conductive organic material to determine both the time course and the final steady-state value. Use of an array of varying non-conductive, semi-conductive, and/or

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conductive organic material thickness would therefore yield
information in a desired fashion.

Accordingly, the invention provides a method for
5   identifying a molecule, the molecule's diffusion
coefficient, the specific activity, structure and/or
function of the molecule.

In one embodiment, the present invention provides a sensor,
10   comprising regions of a first conductive material and a
second material compositionally different than the first
material, wherein the sensor provides an electrical path
through the regions of the first conductive material and
the regions of the second material, wherein the sensor
15   comprises at least one region of second material having a
different thickness than at least one other region of
second material, the second material being selected from
the group consisting of conductive organic material, semi-
conductive material and non-conductive material. The
20   thickness of the second material ranges from about 0.1 µm
to about 100 µm, and typically about 0.1 µm to about 20
µm.

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In another embodiment, the invention provides an array of sensors responsive to a molecule's physical, chemical, or biological characteristics. The array comprises a plurality of sensors, each sensor comprising regions of a

5 first conductive material and a second material compositionally different than the first material, wherein the sensor provides an electrical path through the regions of the first conductive material and the regions of the second material, wherein the sensor comprises at least one

10 region of second material having a different thickness than at least one other region of second material, the second material being selected from the group consisting of conductive organic material, semi-conductive material and non-conductive material.

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The invention provides a broadly responsive analyte detection sensor array based on a variety of "chemiresistor" elements. Such elements are simply prepared and are readily modified chemically to respond to

20 a broad range of analytes. In addition, these sensors yield a rapid, low-power, dc electrical signal in response to the analyte of interest, and their signals are readily integrated with software- or hardware-based algorithms including neural networks for purposes of analyte

identification and physical, biological, chemical

characteristics of the analyte.

In use, the sensors of the invention provide a change in

5 resistance between the conductive elements when contacted

with an analyte or molecule, which interacts second

material (e.g., a material compositionally different than

the first conductive material) of the sensor. The second

material (e.g., a non-conductive material, semi-conductive

10 material or conductive organic material) can be made of any

material designed to interact or bind to a class, genus, or

specie of analyte.

Also provided is a method for determining a physical,

15 chemical, and/or biological characteristics of a molecule.

The method uses a sensing device to produce a

characteristic experimental pattern generated by a

plurality sensors. The pattern has information on the

molecular properties for a molecule or analyte of interest

20 as well as information regarding the analyte's or

molecule's diffusion coefficient data. A response pattern

is produced for each member of the library. The response

patterns may include a change in signal over a period of

time. Such change in the pattern is indicative of the

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diffusion coefficient of a molecule or analyte. These

patterns are then stored and associated with the library.

The library contains patterns for molecules having a

desired or known property or activity.

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In one embodiment, a method is provided for screening

samples for a specific activity or structure by measuring

outputs of a plurality of sensors, each sensor, comprising

regions of a first conductive material and a second

10   material compositionally different than the first material

wherein the sensor comprises at least one region of the

second material having a different thickness than at least

one other region of the second material in the plurality of

sensor, and using results of said measuring to obtain a

15   signal profile, relating to a change in resistance over

time in the plurality of sensors; and comparing the signal

profile to a previously-obtained signal profile indicating

a standard sample having a specific activity, wherein the

signal profile is indicative of a specific activity or a

20   specific structure.

In another embodiment, the invention provides a method of

determining the diffusion coefficient of an analyte,

comprising contacting a sensor with the analtye,

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the sensor comprising, regions of a first conductive

material and a second material compositionally different

than the first material, wherein the sensor provides an

electrical path through the regions of the first material

5   and the regions of the second material, and wherein the

sensor comprises at least one region of second material

having a different thickness than at least one other region

of second material, the second material being selected from

the group consisting of a conductive organic material, a

10 semi-conductive material and a non-conductive material; the

sensors constructed to provide a first response when

contacted with a first chemical analyte, and a second

different response when contacted with a second different

chemical analyte; and measuring a change in the sensor's

15 response to the analyte over time, the change in response

being indicative of the diffusion coefficient of the

analyte.

In another embodiment, the invention provides a method of

20 forming an electrically conductive polymer sensor, said

method comprising, providing a polymer solution comprising

at least a first conductive material and at least a second

material, compositionally different than the first material

in a solvent; providing a substrate; and applying the

polymer solution to the substrate using a spray apparatus.

Brief Description of the Drawing

5   These and other objects of the present invention will now

be described in detail with reference to the accompanying

drawing, in which:

Figure 1A and 1A-1 shows an overview of sensor design; 1B,

10   shows an overview of sensor operation; and 1C, shows an

overview of system operation.

Figure 2 presents the relative differential resistance

responses for various conducting polymer composite sensors

15   to three representative alcohols.

Figure 3 shows a plot of pI_{50} predicted by equation 3 versus

the actual experimental value. Horizontal error bars

represents an average experimental error and vertical error

20   bars correspond to the standard error of equation 3. The

line represents perfect agreement between experiment and

prediction.

Figure 4 shows a diagram illustrating the M and A steric parameters.

Figures 5A-5D shows a table where the first three columns
5   give the name of the alcohol, its experimental pI_{50} value and run in which it was analyzed (and the bubbler in which it was placed). The remainder of the table lists the responses (expressed as percent change in electrical resistance relative to base line resistance) of the 19
10 different polymer/carbon black sensors upon exposure to the alcohols at 5% of their respective saturated vapor pressures. The standard deviation of the responses over ten trials are given in parenthesis. Sensor 14 was not functioning. The last four alcohols were not used in
15 building the model.

Figure 6 illustrates a response of a sensor array of the present invention.

20 Figure 7 illustrates various responses as a function of sensor thickness and analyte volatility.

Detailed Description of the Invention

Studies have shown that certain materials are sensitive to a wide range of gases and vapors and can be used in gas-sensing microelectronic devices. In certain instances, a basic model for such a sensor comprises a thin uniform

5   polymer film lying on top of a pair of coplanar electrodes supported by an insulating substrate. As the gas, or vapor, contact the sensor film it diffuses into the film and is adsorbed at sites randomly distributed throughout the film. The diffusion and adsorption equations can be

10   presented in terms of several fundamental dimensionless parameters that describe the underlying chemical and physical properties of the system. (*see*, Gardner *et al.*, *IEE Proc., Circuits Devices Syst.* **142**, 321-33, (1995)).

15   Accordingly, the invention provides methods of determining a physical, chemical or biological property of a molecule or analyte of interest, including a molecule's or analyte's diffusion coefficient. In one embodiment, the invention measures the molecule's or analyte's interaction with a

20   sensor over a period of time. The measuring can be performed using a parameter proportional to the molecule's concentration in the sample, determining the slope of the linear portion of that relationship, and multiplying that slope by a constant.

Measurements of Diffusion

Sensors that sorb analytes are used in a variety of

detection schemes for sensing the presence of an analyte,

5 for example, an analyte in a fluid. The sorption of

polymers can be transuded into measurable signals through

use of a surface acoustic wave crystal, a quartz

microbalance resonator, a dye-impregnated polymeric coating

on an optical fiber, bulk conducting organic materials, and

10 addition of conductive fillers to insulating organic

polymers to produce a change in electrical resistance of

the composite films. Information about the type and

quantity of an analyte in, for example, a fluid is

contained both in the steady-state response to the presence

15 of an analyte and in the time course of this response as it

approaches its steady-state signal. The ability to obtain

information on the type of, and quantity of, various

analytes in fluids is enhanced by using both the temporal

behavior as well as the steady-state behavior. However,

20 when large numbers of sensors are present, it may not be

feasible or convenient to record enough data points to

obtain the required time course information from each of a

large number of sensors. The need for collection of such

data is obviated by using a collection of nominally

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identical sensors that differ not in composition, but in their thickness or other permeation properties, to the analyte of concern. Accordingly, measurements on the sensors that exhibit rapid permeation to analytes yield

5   steady-state values while measurements at the approximately same time interval on films with poorer analyte permeation yields the desired diffusion coefficient data. In this way it is possible to combine rapid response times on the thinnest films in order to obtain quick information on the

10 presence of an analyte as well as its identity, while simultaneously obtaining kinetic response information that allows measurement of the permeability of the analyte through the film, yielding information on the apparent diffusion coefficient as well as other important kinetic

15 information on the properties of the analyte being detected by the sensors in the array. Thus, through relatively infrequent and slow measurements of the response of an array of such sensors of different thickness, information can be extracted regarding both the diffusion properties

20 and the equilibrium binding properties of the desired analyte, therefore enhancing the performance of vapor detection apparatus.

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A diffusion coefficient indicates the rate at which a molecule moves through a medium under a concentration gradient at a particular temperature and pressure. For example the diffusion of a chemical through a solid

5   particulate is encountered in numerous industrial processes. Diffusion coefficients often must be known to properly design and operate these processes and are useful in determining additional physical characteristics of a sample. For example, in a polyethylene manufacturing

10 process, polymerization occurs in a flammable hydrocarbon solvent such as hexane. After the polymerization, the hexane solvent must be separated and recovered from the polymer to provide a clean resin product. The resin, usually in a form of powder, must be dried to a very low

15 level to minimize the emission of hexane to the environment and the risk of explosion due to hexane build-up in storage vessels. When the hexane in the polyethylene is below 5%, the drying process becomes essentially a process of hexane diffusion in the polymer. The diffusion coefficient

20 therefore is needed to properly design and optimize the process. As another example, crude poly(vinyl chloride) resins usually contain vinyl chloride monomer, a carcinogen. Its diffusion coefficient is needed to

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determine the conditions required to reduce the toxic vinyl

chloride monomer concentration to a safe level.

Most conventional methods of measuring the diffusion

5   coefficient in a plastic material are based on a film

permeability method similar to ASTM D 1434. In such a

method, a film made of the plastic material is placed

between two chambers, one of which holds a constant

concentration of the sample to be tested. The sample

10   permeates through the film into the other chamber and, by

measuring the sample concentration in the second chamber,

one can obtain the diffusion coefficient. Although this

method can produce good precision and accuracy for many

practical applications, it has serious shortcomings if

15   casting a film changes the morphology and physicochemical

characteristics (such as crystallinity) of the material and

if data for unaltered particulates are desired. In

addition, at high temperatures and pressures, the

mechanical integrity of the film may become a problem.

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The diffusion coefficient depends upon the temperature of

the sample. The particles and the diffusion cell can be

heated in an oven and the inert gas should preferably be

heated to the same temperature as the particles.

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As discussed herein, a calculation of the diffusion coefficient is a temporal physical process. Accordingly measurements must be obtained at different time points or

5 during the course of a process of measurement. The two or more measurements give the parameter proportional to the concentration of the molecule in the sample for an interval of time. After a small initial time, the change in the natural log of the parameter per unit time (i.e., the slope

10 of the plot of the natural log of GC area points versus time) will become constant. It is this constant or linear portion of the slope that is used to calculate the diffusion coefficient. The calculation is very simply made by multiplying the slope by a constant. For a spherical or

15 near-spherical particle that constant is $-R^2/\pi^2$ where R is the average radius of a particle. If the particles are in the form of flakes the constant is $-4l^2/\pi^2$, where l is flake thickness, other geometries can be approximated by an equivalent radius using the constant for a spherical

20 particle.

Accordingly, in one embodiment, the invention provides systems, methods and devices for simultaneously determining an equilibrium constant (*i.e.*, partition coefficient) as

well as a diffusion coefficient. In certain instances, a thinner polymer layer is used to determine the partition coefficient and rapid identification of an analyte, whereas a thicker polymer layer can then be used to determine a

5   diffusion coefficient.

The partition coefficient, K, is defined as $K = C_s/C_v$, wherein C_s is the concentration of analyte (solute) in a sorbent phase and C_v is the concentration of the analyte in

10   a vapor phase at equilibrium (at steady-state). In the experimental protocol used herein, C_v is constant since the vapor generation apparatus is continuously replenishing the vapor stream.

15   Using a sensor of the invention, it is possible to concurrently or subsequently determine the diffusion coefficient of an analyte with respect to a particular polymer. As such, the present invention provides a method for measuring a diffusion coefficient of an analyte,

20   comprising: contacting a first sensor having a first predetermined polymer thickness to elicit a first response; contacting a second sensor having a second predetermined polymer thickness to elicit a second response; comparing the first response to the second response to calculate a

time lag and thereafter measuring the diffusion coefficient

of the analyte.

In this aspect, it is assumed that the analyte and the

5   polymer do not react. In the absence of a reaction between

the analyte and the polymer, Fick's Law applies *i.e.*,

$$\frac{\partial C}{\partial t} = D \frac{\partial^2 C}{\partial x^2}$$

Equation I

10   wherein C is the concentration of the analyte and D is the

diffusivity. In order to measure the diffusivity

experimentally in a polymer based sensor, the time lag

procedure is used. Using this procedure, a plot of flux

$(\Delta(\Delta R/R_i)/\Delta t)$ versus times yields a straight line whose

15   intercept θ *i.e.*, the time lag, on the t axis is represented

by Equation II.

$$D = \frac{L^2}{6\theta}$$

Equation II

20   From this graphical relationship the time lag θ is

determined. ΔR is equal to R_t-R_i wherein R_t is the

resistance at time t and R_i is the initial resistance. L is

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the polymer thickness of the sensor (either predetermined

or subsequently determined), the time lag θ is the t

intercept and D is the diffusion coefficient.

5   The diffusion coefficient is an important parameter for a

variety of reasons. For example, by determining the

diffusion coefficient of various polymer and analyte

combinations, it is possible to design a very efficient

sensor system. Thus, the design and efficiency of polymer

10   based sensors depend in part on the diffusion coefficient.

Moreover, the optimum number and kind of polymers in the

sensor array depends in part on the diffusion coefficient.

Thus, if the analyte is an aromatic polar analyte, certain

polymers are better than others. Using the methods of the

15   present invention, optimum polymers can be determined

because the polymer/analyte pair can be better matched.

In certain embodiments, an array of sensors comprise the

same polymer, only the predetermined thickness of the

20   polymer is different. In this aspect, the thinnest polymer

sensor has the most resistance (*i.e.*, fewer conductive

paths to traverse) and the thickest polymer sensor shows

the least resistance (*i.e.*, the most conductive paths to

traverse). However, the thinnest sensor will show steady-

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state conditions faster compared to the thickest sensor

which reach steady state conditions at a slower pace.

Accordingly, where a thickness of a sensor is known one can

obtain merely obtain data at two instances very close in

5   time. For example, with reference to Figure 7, obtaining

data at t=0 and t=0.5 seconds one would be able to

determine the diffusion coefficient based upon the response

of hexane on a "thin" or "thick" film sensor. In this way

the thinner films will come to equilibrium and produce the

10 equilibrium response data even at the short observation

times whereas the thicker films will require longer times

to come to equilibrium and at early time points will

reflect the kinetic response of the sensor to the analyte.

Thus through the use of a combination of thicknesses, one

15 can obtain equilibrium and kinetic response data without

having to record for a lengthy time period on one thick

sensor and wait to equilibrium.

It is important to note that the thickness of the second

20 material of the sensor does not need to be predetermined.

It is helpful, although not essential, to have a

predetermined thickness in providing for ease of

calculating a diffusion coefficient. The thickness may be

calculated subsequent to obtaining data from the

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interaction of the analyte or molecule with at least one

sensor, as described herein. Furthermore, it is not

essential that the polymer thickness be know for purposes

of obtaining a "fingerprint" or "resistance profile" of an

5   analyte. An unknown analyte profile or fingerprint can be

compared to a desired profile or fingerprint to determine

whether the unknown analyte has similar chemical or

biological activity.

10   Sensor Applications

The sensors and sensor arrays disclosed herein act as an

"electronic nose" to offer ease of use, speed, and

identification of analytes and/or analyte regions all in a

portable, relatively inexpensive implementation. Thus, a

15   wide variety of analytes and fluids may be analyzed by the

disclosed sensors, arrays and noses so long as the subject

analyte is capable generating a detectable response across

a plurality of sensors of the array. Analyte applications

include broad ranges of chemical classes such as organics

20   including, for example, alkanes, alkenes, alkynes, dienes,

alicyclic hydrocarbons, arenes, alcohols, ethers, ketones,

aldehydes, carbonyls, carbanions, biogenic amines, thiols,

polynuclear aromatics and derivatives of such organics,

e.g., halide derivatives, etc., biomolecules such as

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sugars, isoprenes and isoprenoids, fatty acids and

derivatives, etc. Accordingly, commercial applications of

the sensors, arrays and noses include environmental

toxicology and remediation, biomedicine, materials quality

5   control, food and agricultural products monitoring,

anaesthetic detection, automobile oil or radiator fluid

monitoring, breath alcohol analyzers, hazardous spill

identification, explosives detection, fugitive emission

identification, medical diagnostics, fish freshness,

10   detection and classification of bacteria and microorganisms

both *in vitro* and *in vivo* for biomedical uses and medical

diagnostic uses, and the like. A wide variety of commercial

applications are available for the sensors arrays and

electronic noses including, but not limited to,

15   environmental toxicology and remediation, biomedicine,

materials quality control, food and agricultural products

monitoring, heavy industrial manufacturing, ambient air

monitoring, worker protection, emissions control, product

quality testing, leak detection and identification, oil/gas

20   petrochemical applications, combustible gas detection, H_2S

monitoring, hazardous leak detection and identification,

emergency response and law enforcement applications,

illegal substance detection and identification, arson

investigation, enclosed space surveying, utility and power

applications, emissions monitoring, transformer fault

detection, food/beverage/agriculture applications,

freshness detection, fruit ripening control, fermentation

process monitoring and control applications, flavor

5   composition and identification, product quality and

identification, refrigerant and fumigant detection,

cosmetic/perfume/fragrance formulation, product quality

testing, personal identification, chemical/plastics/

pharmaceutical applications, leak detection, solvent

10   recovery effectiveness, perimeter monitoring, product

quality testing, hazardous waste site applications,

fugitive emission detection and identification, leak

detection and identification, perimeter monitoring,

transportation, hazardous spill monitoring, refueling

15   operations, shipping container inspection, diesel/gasoline/

aviation fuel identification, building/residential natural

gas detection, formaldehyde detection, smoke detection,

fire detection, automatic ventilation control applications

(cooking, smoking, etc.), air intake monitoring, hospital/

20   medical anesthesia & sterilization gas detection,

infectious disease detection and breath applications, body

fluids analysis, pharmaceutical applications, drug

discovery and telesurgery. Another application for the

sensor-based fluid detection device in engine fluids is an

oil/antifreeze monitor, engine diagnostics for air/fuel

optimization, diesel fuel quality, volatile organic carbon

measurement (VOC), fugitive gases in refineries, food

quality, halitosis, soil and water contaminants, air

5   quality monitoring, leak detection, fire safety, chemical

weapons identification, use by hazardous material teams,

explosive detection, breathalyzers, ethylene oxide

detectors and anesthetics.

10   Biogenic amines such as putrescine, cadaverine, and

spermine are formed and degraded as a result of normal

metabolic activity in plants, animals and microorganisms,

and have been identified and quantified using analytical

techniques such as gas chromatography-mass spectrometry

15   (GC-MS), high performance liquid chromatography (HPLC) or

array based vapor sensing in order to assess the freshness

of foodstuffs such as meats (Veciananogues, 1997, *J. Agr.*

Food Chem., 45:2036-2041), cheeses, alcoholic beverages,

and other fermented foods. Additionally, aniline and o-

20   toluidine have been reported to be biomarkers for patients

having lung cancer (Preti *et al.*, 1988, *J. Chromat. Biomed.*

Appl. 432:1-11), while dimethylamine and trimethylamine

have been reported to be the cause of the "fishy" uremic

breath odor experienced by patients with renal

failure.(Simenhoff, 1977, *New England J. Med.*, 297:132-135)

Thus, in general biogenic amines and thiols are biomarkers

of bacteria, disease states, food freshness, and other

odor-based conditions. Thus, the electronic nose sensors

5   and arrays discussed herein incorporating these materials

can be used to monitor the components in the headspace of

urine, blood, sweat, and saliva of human patients, as well

as breath, to diagnose various states of health and

disease. In addition, they can be used for food quality

10 monitoring, such as fish freshness (which involves volatile

amine signatures), for environmental and industrial

applications (oil quality, water quality, air quality and

contamination and leak detection), for other biomedical

applications, for law enforcement applications

15 (breathalayzers), for confined space monitoring (indoor air

quality, filter breakthrough, etc) and for other

applications delineated above to add functionality and

performance in an unanticipated fashion to existing sensor

arrays though improvement in their properties by use in

20 arrays that combine sensor modalities. For example,

surface acoustic wave (SAW) arrays, quartz crystal

microbalance arrays, composites consisting of regions of

conductors and regions of insulators, bulk semiconducting

organic polymers, and other array types exhibit improved

performance towards vapor discrimination and quantification when the sensors of the present invention are incorporated additionally into arrays that contain these other sensing modalities (e.g., wherein the array of sensors comprises a

5 member selected from the group consisting of a metal oxide gas sensor, a conducting polymer sensor, a dye-impregnated polymer film on fiber optic detector, a polymer-coated micromirror, an electrochemical gas detector, a chemically sensitive field-effect transistor, a carbon black-polymer

10 composite, a micro-electro-mechanical system device and a micro-opto-electro-mechanical system device).

Breath testing has long been recognized as a nonintrusive medical technique that might allow for the diagnosis of

15 disease by linking specific volatile organic vapor metabolites in exhaled breath to medical conditions (see Table 1). Table 1 lists some of the volatile organic compounds that have been identified as targets for specific diseases using gas chromatography/mass spectrometry (GC/MS)

20 methods, with emphasis on amines.

Table 1

Patient Diagnosis	Target VOCs	VOC Source
Uremia; Preti, 1992; Simenhoff, 1977; Davies, 1997	dimethylamine, trimethylamine	breath, urine
Trimethylaminuria; Preti. 1992: Alwaiz.	trimethylamine	breath, urine, swat, vaginal discharge

Preti, 1992; Alwaiz, 1989		vaginal discharge
Lung Cancer; Preti, 1992	aniline, o-toluidine	lung air
Dysgeusia/Dysosmia; Preti, 1992; Oneill, 1988	hydrogen sulfide, methyl mercaptn, pyridine, aniline,diphenylamine, dodecanol	lung air
Cystinuria; Manolis A., 1983, Clin. Chem. 29:5.	cadaverie, piperidine, putrescine, pyrrolidine	breath
Halitosis; Kozlovsky, 1994; Preti, 1992	hydrogen sulfide, methyl mercaptan, cadaverine, putrescine, indole, skatole	mouth air
Bacterial Vaginosis; Chandiok, 1997, J. Clinical Path., 50:790.	amines	vaginal cavity and discharge

With reference now to the drawings, and particularly to FIG. 1, there is shown a sensor array for detecting an analyte in a fluid for use in conjunction with an

5   electrical measuring apparatus. The array comprises a plurality of sensors, at least one of the sensors comprising at least first and second conductive leads electrically coupled to and separated by regions of an a first conductive material and a second material

10   compositionally different than the first conductive material, wherein the sensor provides an electrical path through the regions of the second material and the regions of the conductive material, wherein the sensor comprises at least one region of second material having a different

15   thickness than at least one other region of second material, the second material being selected from the group consisting of conductive organic material, semi-conductive

material and non-conductive or insulating material. The

leads may be any convenient conductive material, usually a

metal, and may be interdigitized to maximize signal-to-

noise strength.

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A sensor in the array is composed of a material comprising

regions of an first conductive material with regions of a

second compositionally dissimilar material. The sensor

comprises a plurality of alternating regions of differing

10 compositions and therefore differing conductivity

transverse to the electrical path between the conductive

leads. Generally, at least one of the sensors is

fabricated by blending a conductive material with an a

material compositionally different than the conductive

15 material. For example, in a colloid, suspension or

dispersion of particulate conductive material in a region

of a second material that is compositionally different, the

regions separating the particles provide changes in

conductance relative to the conductance of the particles

20 themselves. The gaps of different conductance arising from

the dissimilar second material range in path length from

about 10 to 1,000 angstroms, usually on the order of 100

angstroms. The path length and resistance of a given gap

is not constant but rather is believed to change as the

material absorbs, adsorbs or imbibes an analyte.
Accordingly, the dynamic aggregate resistance provided by
these gaps in a given resistor is a function of analyte
permeation of the organic regions of the material. In some
5   embodiments, the conductive material may also contribute to
the dynamic aggregate resistance as a function of analyte
permeation (e.g., when the conductive material is a
conductive organic polymer such as polypyrrole and is
blended with another organic conducting material to form
10   the composite).

A wide variety of conductive materials and dissimilar
second materials can be used. In one embodiment, one such
combination is comprised of an inorganic (Au, Ag) or
15   organic (carbon black) conductive material, while the other
region is comprised of a compositionally dissimilar second
material such as a polymer material (e.g., polyaniline,
polypyrrole, polythiophene, polyEDOT, and other conducting
organic polymers such as those in the Handbook of
20   Conducting Polymers (Handbook of Conducting Polymers,
second ed., Marcel Dekker, New York 1997, vols. 1 & 2)).
Other combinations of conductor/dissimilar second
material/composite materials are also useful.

30

Polyaniline is a desirable member in the class of conducting organic polymer materials in that the half oxidized form, the emeraldine base, is rendered electrically conductive upon incorporation of a strong

5   acid. The conductive form of polyaniline, commonly referred to as the emeraldine salt (ES), has been reported to deprotonate to the emeraldine base and become insulating in alkaline environments. Without being bound to any particular theory, the polyaniline may also undergo a

10 beneficial phase transition that also contributes to the superior performance of such composites.

Table 2 provides exemplary conductive materials for use in sensor fabrication; blends, such as of those listed, may

15 also be used. Typically conductors include, for example, those having a positive temperature coefficient of resistance. The sensors are comprised of a plurality of alternating regions of a conductor with regions of a compositionally dissimilar material. Without being bound

20 to any particular theory, it is believed that an electrical charge traverses between the two contacting electrodes traverses both the regions of the conductor and the regions of the second dissimilar material.

Table 2:

Major Class	Examples
Organic Conductors	conducting polymers (poly(anilines), poly(thiophenes), poly(pyrroles), poly(aceylenes, *etc.*)), carbonaceous material (carbon blacks, graphite, coke, C60 *etc.*), charge transfer complexes (tetramethylparaphenylenediamine-chloranile, alkali metal tetracyanoquinodimethane complexes, tetrathiofulvalene halide complexes, *etc.*), *etc.*
Inorganic Conductors	metals and metal alloys (Ag, Au, Cu, Pt, AuCu alloy, *etc.*), highly doped semiconductors (Si, GaAs, InP, MoS2, TiO2, *etc.*), conductive metal oxides (In2O3, SnO2, Na2Pt3O4, *etc.*), superconductors (Yba2Cu3O7, Ti2Ba2Ca2Cu3O10, *etc.*), *etc.*
Mixed inorganic/organic Conductor	Tetracyanoplatinate complexes, Iridium halocarbonyl complexes, stacked macrocyclic complexes. *Etc.*

In this embodiment, the conducting region can be anything that can carry electrons from atom to atom, including, but not limited to, a material, a particle, a metal, a polymer,

5 a substrate, an ion, an alloy, an organic material, (e.g., carbon, graphite, etc.) an inorganic material, a biomaterial, a solid, a liquid, a gas or regions thereof.
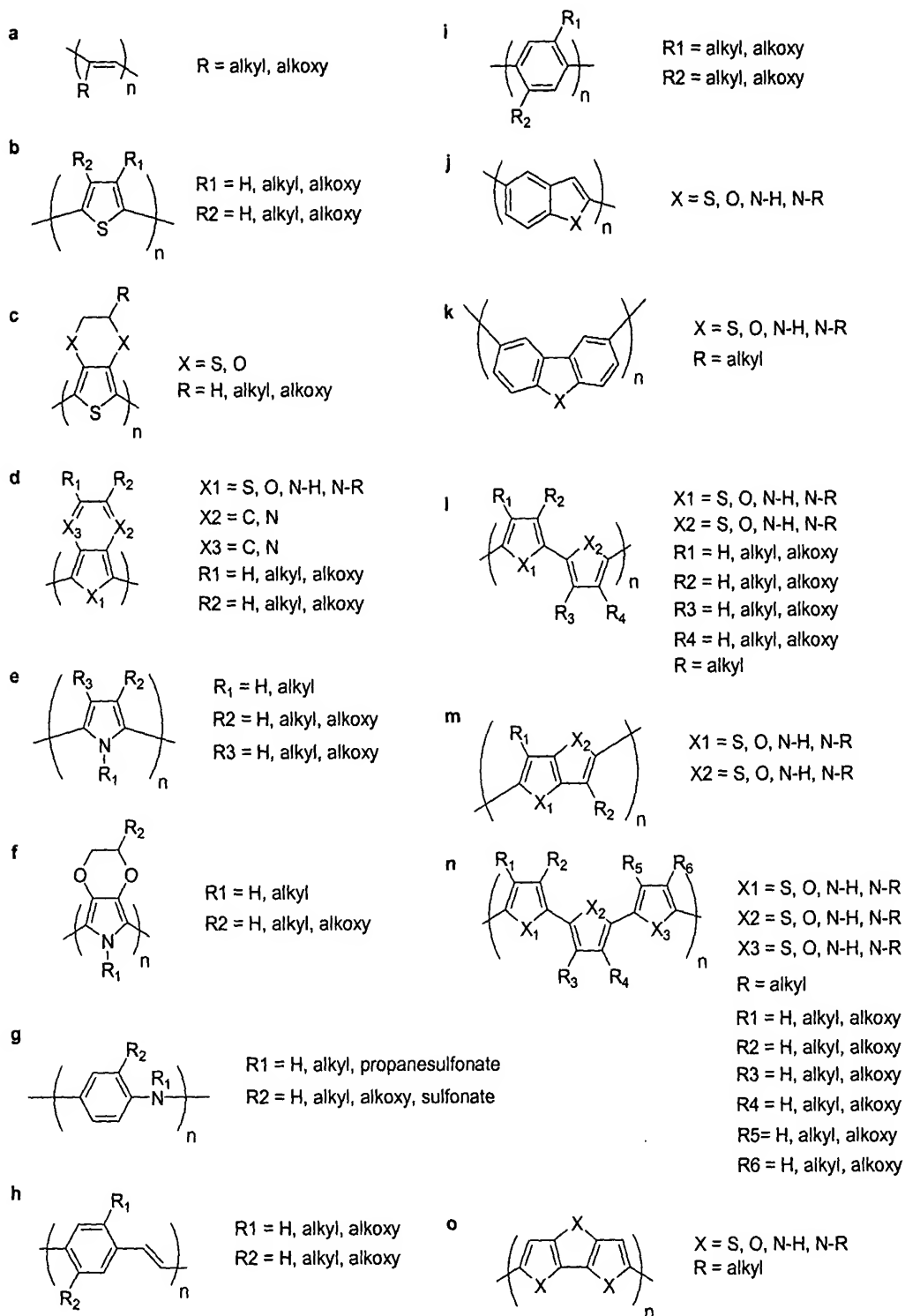
In certain other embodiments, the conductive material is a

10 conductive particle, such as a colloidal nanoparticle. As used herein the term "nanoparticle" refers to a conductive cluster, such as a metal cluster, having a diameter on the nanometer scale. Such nanoparticles are optionally stabilized with organic ligands.
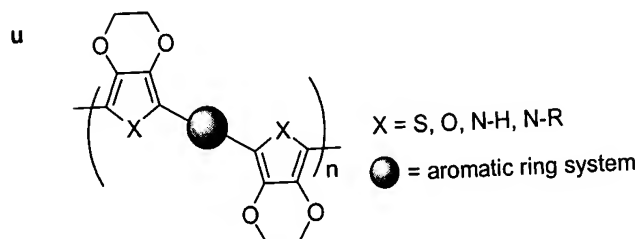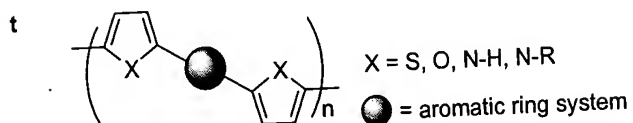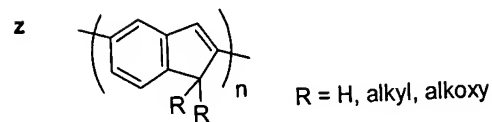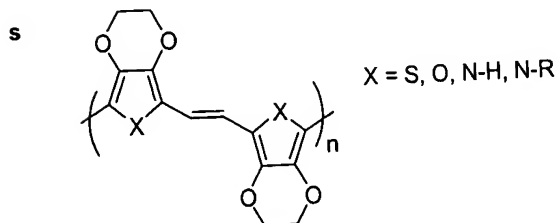
32

Examples of colloidal nanoparticles for use in accordance
with the present invention are described in the literature.
In this embodiment, the organic region can optionally be a

5   ligand that is attached to a central core making up the
nanoparticle. These ligands *i.e.*, caps, can be polyhomo-
or polyhetero-functionalized, thereby being suitable for
detecting a variety of chemical analytes. The
nanoparticles, *i.e.*, clusters, are stabilized by the

10   attached ligands. In certain embodiments, the conducting
component of the resistors are nanoparticles comprising a
central core conducting element and an attached ligand
optionally in a polymer matrix. With reference to Table 2,
various conducting materials are suitable for the central
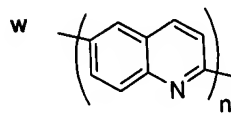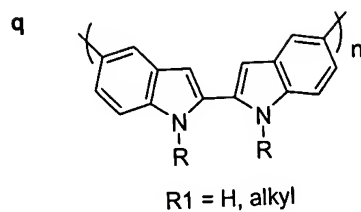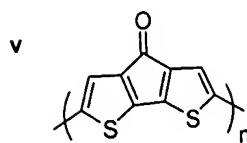
15   core. In certain embodiments, the nanoparticles have a
metal core. Typcial metal cores include, but are not
limited to, Au, Ag, Pt, Pd, Cu, Ni, AuCu and regions
thereof. Gold (Au) is especially preferred. These
metallic nanoparticles can be synthesized using a variety

20   of methods. In a preferred method of synthesis, a
modification of the protocol developed by Brust *et al.* can
be used. (*see*, Brust, M.; Walker, M.; Bethell, D.;
Schiffrin, D.J.; Whyman, R. *J. Chem. Soc., Chem. Commun.*,
1994, 801-802.) As explained more fully below, by varying

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the concentration of the synthetic reagents, the particle

size can be manipulated and controlled.

Table 3 provides exemplary conductive organic materials

5   that can be used to form a the second dissimilar material

regions of the sensors.

TABLE 3:

a

R = alkyl, alkoxy

b

R1 = H, alkyl, alkoxy
R2 = H, alkyl, alkoxy

c

X = S, O
R = H, alkyl, alkoxy

d

X1 = S, O, N-H, N-R
X2 = C, N
X3 = C, N
R1 = H, alkyl, alkoxy
R2 = H, alkyl, alkoxy

e

R1 = H, alkyl
R2 = H, alkyl, alkoxy
R3 = H, alkyl, alkoxy

f

R1 = H, alkyl
R2 = H, alkyl, alkoxy

g

R1 = H, alkyl, propanesulfonate
R2 = H, alkyl, alkoxy, sulfonate

h

R1 = H, alkyl, alkoxy
R2 = H, alkyl, alkoxy

i

R1 = alkyl, alkoxy
R2 = alkyl, alkoxy

j

X = S, O, N-H, N-R

k

X = S, O, N-H, N-R
R = alkyl

l

X1 = S, O, N-H, N-R
X2 = S, O, N-H, N-R
R1 = H, alkyl, alkoxy
R2 = H, alkyl, alkoxy
R3 = H, alkyl, alkoxy
R4 = H, alkyl, alkoxy
R = alkyl

m

X1 = S, O, N-H, N-R
X2 = S, O, N-H, N-R

n

X1 = S, O, N-H, N-R
X2 = S, O, N-H, N-R
X3 = S, O, N-H, N-R
R = alkyl
R1 = H, alkyl, alkoxy
R2 = H, alkyl, alkoxy
R3 = H, alkyl, alkoxy
R4 = H, alkyl, alkoxy
R5 = H, alkyl, alkoxy
R6 = H, alkyl, alkoxy

o

X = S, O, N-H, N-R
R = alkyl

35

p

R1 = H, alkyl, alkoxy
R2 = H, alkyl, alkoxy

v

q

R1 = H, alkyl

w

x

r

X = S, O, N-H, N-R
R1 = H, alkyl, alkoxy
R2 = H, alkyl, alkoxy

y

R = H, alkyl, alkoxy

s

X = S, O, N-H, N-R

z

R = H, alkyl, alkoxy

t

X = S, O, N-H, N-R
= aromatic ring system

u

X = S, O, N-H, N-R
= aromatic ring system

a. Poly(acetylene) and derivatives
b. Poly(thiophenes) and derivatives

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c.   Poly(3,4-ethylenedioxythiophene) and poly(3,4-
ethylenedithiathiophene)   and derivatives
d.   Poly(isathianaphthene), poly(pyridothiophene),
poly(pyrizinothiophene),   and derivatives
5   e.   Poly(pyrrole) and derivatives
f.   Poly(3,4-ethylenedioxypyrrole) and derivatives
g.   Poly(aniline) and derivatives
h.   Poly(phenylenevinylene) and derivatives
I.   Poly(p-phenylene) and derivatives
10   j.   Poly(thianapthene), poly(benxofuran), and poly(indole) and
derivatives
k.   Poly(dibenzothiophene), poly(dibenxofuran), and poly(carbazole)
and   derivatives
l.   Poly(bithiophene), poly(bifuran), poly(bipyrrole), and
15   derivatives
m.   Poly(thienothiophene), poly(thienofuran), poly(thienopyrrole),
poly(furanylpyrrole), poly(furanylfuran),
poly(pyrolylpyrrole), and   derivatives
n.   Poly(terthiophene), poly(terfuran), poly(terpyrrole), and
20   derivatives
o.   Poly(dithienothiophene), poly(difuranylthiophene),
poly(dipyrrolylthiophene), poly(dithienofuran),
poly(dipyrrolylfuran), poly(dipyrrolylpyrrole) and derivatives
p.   Poly(phenyl acetylene) and derivatives
25   q.   Poly(biindole) and derivatives
r.   Poly(dithienovinylene), poly(difuranylvinylene), poly
(dipyrrolylvinylene) and derivatives
s.   Poly(1,2-trans(3,4-ethylenedioxythienyl)vinylene), poly(1,2-
trans(3,4-ethylenedioxyfuranyl)vinylene), and poly(1,2-trans(3,4-
30   ethylenedioxypyrrolyl)vinylene), and derivatives
t.   The class of poly(bis-thienylarylenes) and poly(bis-
pyrrolylarylenes) and derivatives
u.   The class of poly(bis(3,4-ethylenedioxythienyl)arylenes) and
derivatives
35   v.   Poly(dithienylcyclopentenone)
w.   Poly(quinoline)
x.   Poly(thiazole)
y.   Poly(fluorene) and derivatives
z.   Poly(azulene) and derivatives
40   **Notes:**
a.   Aromatics = phenyl, biphenyl, terphenyl, carbazole, furan,
thiophene, pyrrole, fluorene, thiazole, pyridine, 2,3,5,6-
hexafluorobenzene, anthracene, coronene, indole, biindole, 3,4-
ethylenedioxythiophene, 3,4-ethylenedioxypyrrole, and both the
45   alkyl and alkoxy derivatives of these aromatics.
b.   Alkyl = aliphatic group branched or straight chain ranging from
CH_3 to $C_{20}H_{41}$.
c.   Alkoxy = OR, where R is an aliphatic group that may either be
branched or straight chain ranging from CH_3 to $C_{20}H_{41}$.
50   d.   All conductive polymers are depicted in their neutral,
nonconductive form. The polymers listed in the figure are doped
oxidatively either by means chemically or electrochemically.
e.   The class of polyanilines are acid doped and can be done so with
a number of sulfonic acids including methane sulfonic acid,
55   ethane sulfonic acid, propane sulfonic acid, butane sulfonic
acid, pentane sulfonic acid, hexane sulfonic acid, heptane
sulfonic acid, octane sulfonic acid, nonane sulfonic acid, decane

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sulfonic acid, ondecane sulfonic acid, dodecane sulfonic acid, dodecylbenzenesulfonic acid, toluene sulfonic acid, benzene sulfonic acid, dinonanylnaphthalene sulfonic acid, and both the d and 1 forms of camphor sulfonic acid.

5   f.   All other class of conductive polymers when doped there is an associated counter ion to compensate the positive charges on the backbone. These can be perchlorate, hexafluorophosphate, tetrafluoroborate, fluoride, chloride, bromide, iodide, triflate, etc.

10

The organic material can be either an organic semiconductor or organic conductor. "Semi-conductors" as used herein, include materials whose electrical conductivity increases as the temperature increases, whereas conductors are

15   materials whose electrical conductivity decreases as the temperature increases. By this fundamental definition, the organic materials that are useful in the sensors of the present invention are either semiconductors or conductors. Such materials are collectively referred to herein as

20   organic materials because they produce a readily-measured resistance between two conducting leads separated by about 10 micron or more using readily-purchased multimeters having resistance measurement limits of 100 Mohm or less, and thus allow the passage of electrical current through

25   them when used as elements in an electronic circuit at room temperature. Semi-conductors and conductors can be differentiated from insulators by their different room temperature electrical conductivity values. Insulator show very low room temperature conductivity values, typically

less than about 10^{-8} ohm^{-1} cm^{-1}. Poly(styrene),

poly(ethylene), and other polymers elaborated in Table 4

provide examples of insulating or "non-conductive"

materials. Metals have very high room temperature

5   conductivities, typically greater than about 10 ohm^{-1} cm^{-1}.

Semi-conductors have conductivities greater than those of

insulators, and are distinguished from metals by their

different temperature dependence of conductivity, as

described above. Examples of semi-conducting and

10   conducting organic material are provided in Table 3. The

organic materials that are useful in the sensors of the

present invention are either semiconductors or conductors,

and have room temperature electrical conductivities of

greater than about 10^{-6} ohm^{-1} cm^{-1}, preferably having a

15   conductivity of greater than about 10^{-3} ohm^{-1} cm^{-1}.

Accordingly, the sensors of the present invention include

sensors comprising regions of an electrical conductor and

regions of a compositionally different material that is an

20   electrical conductor, semiconductor, or non-conductive

material. As used above, electrical conductors include,

for example, Au, Ag, Pt and carbon black, other conductive

materials having similar resistivity profiles are easily

identified in the art (see, for example the latest edition

of: *The CRC Handbook of Chemistry and Physics*, CRC Press,

the disclosure of which is incorporated herein by

reference). Furthermore, non-conductive materials (i.e.,

insulators) can also be incorporated into the composite to

5   further manipulate the analyte response properties of the

composites. The insulating region (i.e., non-conductive

region) can be anything that can impede electron flow from

atom to atom, including, but not limited to, a polymer, a

plasticizer, an organic material, an organic polymer, a

10   filler, a ligand, an inorganic material, a biomaterial, a

solid, a liquid, a gas and combinations thereof.   Table 4

provides examples of a non-conductive second material

(i.e., a second material that is compositionally different

than the conductive material of a sensor) that can be used

15   for such purposes.

TABLE 4:

Major Class	Examples
Main-chain carbon polymers	poly(dienes), poly(alkenes), poly(acrylics), poly(methacrylics), poly(vinyl ethers), poly(vinyl thioethers), poly(vinyl alcohols), poly(vinyl ketones), poly(vinyl halides), poly(vinyl nitrites), poly(vinyl esters), poly(styrenes), poly(aryines), *etc.*
Main-chain acyclic heteroatom polymers	poly(oxides), poly(caronates), poly(esters), poly(anhydrides), poly(urethanes), poly(sulfonate), poly(siloxanes), poly(sulfides), poly(thioesters), poly(sulfones), poly(sulfonamindes), poly(amides),

	poly(ureas), poly(phosphazens), poly(silanes), poly(silazanes), *etc.*
Main-chain heterocyclic polymers	poly(furantetracarboxylic acid diimides), poly(benzoxazoles), poly(oxadiazoles), poly(benzothiazinophenothiazines), poly(benzothiazoles), poly(pyrazinoquinoxalines), poly(pyromenitimides), poly(quinoxalines), poly(benzimidazoles), poly(oxidoles), poly(oxoisinodolines), poly(diaxoisoindoines), poly(triazines), poly(pyridzaines), poly(pioeraziness), poly(pyridinees), poly(pioeridiens), poly(triazoles), poly(pyrazoles), poly(pyrrolidines), poly(carboranes), poly(oxabicyclononanes), poly(diabenzofurans), poly(phthalides), poly(acetals), poly(anhydrides), carbohydrates, *etc.*

Nonconductive organic polymer materials; blends and copolymers; plasticized polymers; and other variations including those using the polymers listed here, may also be

5   used. Combinations, concentrations, blend stoichiometries, percolation thresholds, etc. are readily determined empirically by fabricating and screening prototype resistors (chemiresistors) as described below.

10   <u>Sensor Fabrication</u>

The sensors of the invention can be fabricated by many techniques such as, but not limited to, solution casting, suspension casting, air-brush techniques and mechanical mixing.

41

The sensors as described above can be fabricated by using a standard painter's air-brush (or equivalents easily recognized in the art). For example, an air-brush can be

5 used to coat a substrate with a mixture of any number of various combinations of the foregoing conductive materials and a second material that is compositionally different than the conductive material.

10 In one embodiment, a polymer solution or suspension is placed in a reservoir and the reservoir attached to a compressed gas system. The flowing gas sucks up the solution or suspension by a vacuum created in the air brush system, which is then ejected from a nozzle of the air

15 brush. This flow of solution or suspension (e.g., polymer, solvent and suspended material) is directed toward a substrate that has been prepared to receive the polymer solution or suspension. The polymer solution or suspension can be stirred either continuously or intermittently while

20 in the reservoir in order to maintain a homogenous mixture.

The method of the invention is amenable to automation using, for example, a computer system to control the rate and amount of polymer solution or suspension ejected from a

nozzle of the air brush system. Accordingly, it will be recognized that the method of the invention is capable of fabricating many sensors in the time it would take to make one using the standard methods of dip coating or spin

5 coating. Additionally, the method of the invention is capable of high reproducibility in sensor fabrication.

It will also be recognized that the method of the invention makes it possible to simply and efficiently fabricate

10 sensors from various and odd shaped substrates. For example, the substrates need not be flat as required by standard spin coating techniques.

The substrates can be prepared such that a large number of

15 substrates are contacted by a single pass of the air-brush system. This leads to a large number of very reproducible sensors (determined by baseline resistance) in a simple and time efficient process.

20 In addition, the method of the invention lends itself to making simple changes in sensor thickness and sensor compositions. For example, if a sensor is desired that has a thicker polymer coating than another sensor, the system merely ejects more polymer solution or suspension or ejects

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for a longer period of time at a particular location in an array of sensors or on a particular sensor.

In addition, the methods of the invention could be used to

5   fabricate sensors having small wells or specific regions on an otherwise smooth surface. The method involves ejecting a solution or suspension of polymer as a fine jet or stream directed to a particular location on a substrate or sensor. In other words, the spray jet could be made to pass over

10   the whole substrate (as described above) or directed at only a small area thus containing the coated region without the need for mask to prevent coating or non-desired regions. Accordingly, depending on the type of application required custom nozzles that provide custom jets or streams

15   of polymer could be designed. For example, nozzles that provide conical sprays, linear sprays, planer sprays, or other spray geometries can be designed to those of skill in the art. A number of substrates may be employed for this purpose including, but not limited to, those formed of

20   metals, organic polymers, inorganic polymers, ceramics, textiles, and composites thereof.

For systems where the conducting and/or the compositionally dissimilar material or their reaction precursors are

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soluble in a common solvent, the sensor (e.g., the

chemiresistor) can be fabricated by solution casting. The

oxidation of pyrrole by phosphomolybdic acid represents

such a system. In this reaction, the phosphomolybdic acid

5   and pyrrole are dissolved in tetrahydrofuran (THF) and

polymerization occurs upon solvent evaporation. For

example, this method allows for THF soluble compositionally

different conductive, semiconductive, and non-conductive

materials to be dissolved into this reaction region thereby

10   allowing the composite to be formed in a single step upon

solvent evaporation.

Certain conducting organic polymers can also be synthesized

via a soluble precursor polymer. In these cases, blends

15   between the precursor polymer and the compositionally

different material of the composite can first be formed

followed by chemical reaction to convert the precursor

polymer into the desired conducting polymer. For instance

poly(p-phenylene vinylene) can be synthesized through a

20   soluble sulfonium precursor. Blends between this sulfonium

precursor and a non-conductive or conductive polymer can be

formed by solution casting. After which, the blend can be

subjected to thermal treatment under vacuum to convert the

sulfonium precursor to the desired poly(p-phenylene vinylene).

In suspension casting, one or more of the components of the
5 sensor is suspended and the others dissolved in a common solvent. Suspension casting is a rather general technique applicable to a wide range of species, such as carbon blacks or colloidal metals, which can be suspended in solvents by vigorous mixing or sonication. In one
10 application of suspension casting, the organic or conductive polymer is dissolved in an appropriate solvent (such as THF, acetonitrile, water, etc.). Carbon black is then suspended in this solution and the resulting region is used to dip coat or spray coat electrodes.
15

Mechanical mixing is suitable for all of the conductive/ conductive organic/non-conductive combinations possible. In this technique, the materials are physically mixed in a ball-mill or other mixing device. For instance, carbon
20 black/conducting organic polymer composites are readily made by ball-milling. When the semi-conductive or conductive organic material can be melted or significantly softened without decomposition, mechanical mixing at elevated temperature can improve the mixing process.

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Alternatively, composite fabrication can sometimes be improved by several sequential heat and mix steps.

Once fabricated, the individual sensors can be optimized for a particular application by varying their chemical make up and morphologies. The chemical nature of the sensors determines to which analytes they will respond and their ability to distinguish different analytes. The relative ratio of conductive to compositionally different material (e.g., non-conductive, semi-conductive material), along with the composition of any other insulating organic or inorganic components, can determine the magnitude of the response since the resistance of the elements becomes more sensitive to sorbed molecules as the percolation threshold is approached and as the molecules interact chemically with the components of the composite that adsorb or absorb the analyte. The film morphology is also important in determining response characteristics. For instance, uniform thin films respond more quickly to analytes than do uniform thick ones. Hence, with an empirical catalogue of information on chemically diverse sensors made with varying ratios of semiconductive, conducting, and insulating components and by differing fabrication routes, sensors can be chosen that are appropriate for the analytes expected in

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a particular application, their concentrations, and the desired response times. Further optimization can then be performed in an iterative fashion as feedback on the performance of an array under particular conditions becomes

5   available. As mentioned above, by varying the thickness of, for example, the organic material of the sensor it is possible to obtain information regarding the diffusion characteristics of a certain analyte or molecule of interest.

10

Sensor arrays particularly well-suited to scaled up production are fabricated using integrated circuit (IC) design technologies. For example, the sensor can easily be integrated onto the front end of a simple amplifier

15   interfaced to an A/D converter to efficiently feed the data stream directly into a neural network software or hardware analysis section. Micro-fabrication techniques can integrate the sensor directly onto a micro-chip which contains the circuitry for analog signal

20   conditioning/processing and then data analysis. This provides for the production of millions of incrementally different sensor elements in a single manufacturing step using, for example, ink-jet technology.

In one embodiment, the sensor arrays have a predetermined inter-sensor variation in the structure, thickness or composition of the first conductive material or the second compositionally different materials as well as in the

5 conductive components and any insulating or plastizing components of the composites. The variation may be quantitative and/or qualitative. For example, the concentration of the conductive or semi-conductive organic material in the composite can be varied across sensors.

10 Alternatively, a variety of different organic materials may be used in different sensors. This ability to fabricate many chemically different materials allows ready incorporation of a wide range of chemical diversity into the sensor elements, and also allows facile control over

15 the electrical properties of the sensor elements through control over the composition of an individual sensor element in the array. Insulating organic materials can also be used and blended into the array in order to further increase the diversity in one embodiment of the invention.

20 Commercially available, off-the-shelf, organic polymers can provide the basic sensor components that respond differently to different analytes, based on the differences in polarity, molecular size, and other properties of the analyte in order to achieve the chemical diversity amongst

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array elements in the electronic nose sensors. Such

diversity can be further enhanced by varying the thickness

of a sensor or region of a sensor. Otherwise, these

properties can be obtained by modification in the

5   composition of the electrically conductive or reactive

second material component of the sensor composition by use

of capping agents on a colloidal metal part of the

conductive phase, by use of different plasticizers added to

otherwise compositionally identical sensor elements to

10   manipulate their analyte sorption and response properties,

by variation in the temperature or measurement frequency of

the sensors in an array of sensors that are otherwise

compositionally identical, or a combination thereof and

with sensors that are compositionally different as well.

15   The sensors in an array can readily be made by

combinatorial methods in which a limited number of

feedstocks are combined to produce a large number of

chemically distinct sensor elements.

20   As used herein the term "predetermined" means that the

conductive film layer associated with the sensor has, for

example, a predefined or predetermined thickness which is

measured within about 1 nm using atomic force microscopy,

scanning tunneling microscopy, profilometry or other
suitable method known by those of skill in the art.

One method of enhancing the diversity of polymer based
5 conductor/conductor, conductor/semiconductor
conductor/insulator, sensors and combinations thereof is
through the use of polymer blends or copolymers (Doleman,
et al. (1998) Anal. Chem. 70, 2560-2654). Immiscible
polymer blends may also be of interest because carbon black
10 or other conductors can be observed to preferentially
segregate into one of the blend components.

Sensor arrays allow expanded utility because the signal for
an imperfect "key" in one channel can be recognized through
15 information gathered on another, chemically or physically
dissimilar channel in the array. A distinct pattern of
responses produced over the collection of sensors in the
array can provide a fingerprint that allows classification
and identification of the analyte, whereas such information
20 would not have been obtainable by relying on the signals
arising solely from a single sensor or sensing material.
In addition, by varying the thickness of regions or whole
sensors a distinctive pattern may be further expanded to
include data on the diffusion coefficient of a particular

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molecule or analyte. The data may be obtained through the measurement of a single sensor, which measures the diffusion through a certain organic material (e.g., a semi-conductive or non-conductive material) or through the

5   diffusion of a number of dissimilar materials.

The general method for using the disclosed sensors, arrays and electronic noses, for detecting the presence of an analyte in a fluid, where the fluid is a liquid or a gas,

10   involves resistively sensing the presence of an analyte in a fluid with a chemical sensor comprising first and second conductive leads electrically coupled to and separated by a chemically sensitive sensor as described above by measuring a first resistance between the conductive leads when the

15   resistor is contacted with a first fluid comprising first analyte and a second, different resistance when the resistor is contacted with a second, different fluid. Where a diffusion coefficient is to be determined a resistivity at a first time when contacted with the analyte

20   is measured followed by a measurement of resistivity at a second time. If the measurements are by analog signal the measurements may be obtained continuously during the time period for measuring the diffusion of the analyte. Where

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an analog to digital converter is present measurements that
are essentially continuous can be obtained.

An ideal detector array would produce a unique signature
5   for every different analyte to which it was exposed. To
construct such a system, it is necessary to include
detectors that probe important, but possibly subtle,
molecular parameters such as, for example chirality. The
term "chiral" is used herein to refer to an optically
10 active or enantiomerically pure compound, or to a compound
containing one or more asymmetric centers in a well-defined
optically active configuration. A chiral compound is not
superimposable upon its mirror image. Harnessing
enantiomer resolution gives rise to myriad applications.
15 For instance, because the active sites of enzymes are
chiral, only the correct enantiomer is recognized as a
substrate. Thus, pharmaceuticals having near enantiomeric
purity are often many more times active than their racemic
mixtures. However, many pharmaceutical formulations
20 marketed today are racemic regions of the desired compound
and its "mirror image." One optical form (or enantiomer)
of a racemic region may be medicinally useful, while the
other optical form may be inert or even harmful, as has
been reported to be the case for thalidomide. In this

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fashion, the sensors and sensor arrays would be useful in assessing which form of chirality, and of what enantiomeric excess, was present in an analyte in a fluid. Due to the presence of chiral moieties, many biomolecules, such as

5   amino acids, are amenable to detection using the sensor arrays of the present invention.

Similarly, by characterizing the diffusion characteristics of a molecule or analyte, it is possible to screen for

10   molecules or analytes having diffusion coefficients similar or identical to a desired diffusion coefficient. This is useful in identifying molecules useful as pharmaceutical. For example, a beneficial drug or biological agent of interest may demonstrate a particular binding affinity for

15   a polymer material of interest. This same biological agent may have better efficacy due to its diffusion through biological membranes. Accordingly, screening combinatorial drugs for biological agents using the sensors of the invention can measure not only the biological agent's

20   binding affinity but its chirality and diffusion coefficient.

In another embodiment, the sensor for detecting the presence of a chemical analyte in a fluid comprises a

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sensor electrically coupled to an electrical measuring

apparatus where the sensor is in thermal communication with

a temperature control apparatus. As described above, the

sensor comprises regions of a first conductive material and

5   regions of a second material that is is compositionally

different than the first conductive material. The sensor

provides an electrical path through which electrical

current may flow and a resistance (R) at a temperature (T)

when contacted with a fluid comprising a chemical analyte.

10

In operation, the sensor(s) for detecting the presence of a

chemical analyte in a fluid provide an electrical

resistance (R_m) when contacted with a fluid comprising a

chemical analyte at a particular temperature (T_m). The

15   electrical resistance observed may vary as the temperature

varies, thereby allowing one to define a unique profile of

electrical resistances at various different temperatures

for any chemical analyte of interest. In addition, the

profile will include data characteristic of the diffusion

20   coefficient of the analyte of interest. The diffusion

coefficient will also be affected by the temperature as

discussed above. For example, a chemically sensitive

resistor, when contacted with a fluid comprising a chemical

analyte of interest, will provide an electrical resistance

R_m at temperature T_m where m is an integer greater than 1, and may provide a different electrical resistance R_n at a different temperature T_n. The difference between R_m and R_n is readily detectable by an electrical measuring apparatus.

5 If the sensor comprises an array of two or more sensors each being in thermal communication with a temperature control apparatus, one may vary the temperature across the entire array (i.e., generate a temperature gradient across the array), thereby allowing electrical resistances to be

10 measured simultaneously at various different temperatures and for various different resistor compositions. For example, in an array of sensors, one may vary the composition of the sensors in the horizontal direction across the array, such that sensor composition in the

15 vertical direction across the array remains constant. One may then create a temperature gradient in the vertical direction across the array, thereby allowing the simultaneous analysis of chemical analytes at different sensor compositions and different temperatures.

20

Methods for placing chemically sensitive resistors in thermal communication with a temperature control apparatus are readily apparent to those skilled in the art and include, for example, attaching a heating or cooling

element to the sensor and passing electrical current through said heating or cooling element.

In yet another embodiment, rather than subjecting the
5 sensor to a direct electrical current and measuring the true electrical resistance through the sensor(s) can be subjected to an alternating electrical current at different frequencies to measure impedance. Impedance is the apparent resistance in an alternating electrical current as compared
10 to the true electrical resistance in a direct current. As such, the present invention is also directed to a sensor for detecting the presence of a chemical analyte in a fluid, said sensor connected to an electrical measuring apparatus, the sensor comprising regions of a first
15 conductive material and a second material compositionally different than the first conductive material and wherein said sensor provides (a) an electrical path through said region of organic material and said conductive material, and (b) an electrical impedance Z_m at frequency m when
20 contacted with a fluid comprising an analyte, where m is an integer greater than 1 and m does not equal 0. One skilled in the art will recognize that variations in thickness of the sensor materials can be implemented to measure a diffusion coefficient. For measuring impedance as a

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function of frequency, the frequencies employed will

generally range from about 1 Hz to 5 GHz, usually from

about 1 MHZ to 1 GHz, more usually from about 1 MHZ to 10

MHZ and preferably from about 1 MHZ to 5 MHZ. An analyte

5 will exhibit unique impedance characteristics at varying

alternating current frequencies, thereby allowing one to

detect the presence of any chemical analyte of interest in

a fluid by measuring Z_m at alternating frequency m.

Similarly, the impedance will vary according to the

10 diffusion of the analyte in the sensor.

For performing impedance measurements, one may employ

virtually any impedance analyzer known in the art. For

example, a Schlumberger Model 1260 Impedance/Gain-Phase

15 Analyzer (Schlumberger Technologies, Farmborough,

Hampshire, England) with approximately 6 inch RG174 coaxial

cables is employed. In such an apparatus, the resistor/

sensor is held in an Al chassis box to shield it from

external electronic noise.

20

In still another embodiment of the present invention, one

may vary both the frequency m of the electrical current

employed and the temperature T_n and measure the electrical

impedance $Z_{m,n}$, thereby allowing for the detection of the presence of a chemical analyte of interest.

An electronic nose for detecting an analyte in a fluid is
5   fabricated by electrically coupling the sensor leads of an array of sensors to an electrical measuring device. The device measures changes in signal at each sensor of the array, preferably simultaneously and preferably over time. Preferably, the signal is an electrical resistance,
10   although it could also be an impedance or other physical property of the material in response to the presence of the analyte in the fluid. Frequently, the device includes signal processing means and is used in conjunction with a computer and data structure for comparing a given response
15   profile to a structure-response profile database for qualitative and quantitative analysis. Typically such a nose comprises usually at least ten, often at least 100, and perhaps at least 1000 different sensors though with mass deposition fabrication techniques described herein or
20   otherwise known in the art, arrays of on the order of at least one million sensors are readily produced.

In one embodiment, the temporal response of each sensor (resistance as a function of time) is recorded. The

temporal response of each sensor may be normalized to a

maximum percent increase and percent decrease in signal

which produces a response pattern associated with the

exposure of the analyte. By iterative profiling of known

5   analytes, a structure-function database correlating

analytes and response profiles is generated. Unknown

analytes may then be characterized or identified using

response pattern comparison and recognition algorithms.

Accordingly, analyte detection systems comprising sensor

10   arrays, an electrical measuring device for detecting

resistance across each chemiresistor, a computer, a data

structure of sensor array response profiles, and a

comparison algorithm are provided. In another embodiment,

the electrical measuring device is an integrated circuit

15   comprising neural network-based hardware and a digital-

analog converter (DAC) multiplexed to each sensor, or a

plurality of DACs, each connected to different sensor(s).

The desired signals if monitored as dc electrical

20   resistances for the various sensor elements in an array can

be read merely by imposing a constant current source

through the resistors and then monitoring the voltage

across each resistor through use of a commercial

multiplexable 20 bit analog-to-digital converter. Such

60

signals are readily stored in a computer that contains a

resident algorithm for data analysis and archiving.

Signals can also be preprocessed either in digital or

analog form; the latter might adopt a resistive grid

5   configuration, for example, to achieve local gain control.

In addition, long time adaptation electronics can be added

or the data can be processed digitally after it is

collected from the sensors themselves. This processing

could be on the same chip as the sensors but also could

10   reside on a physically separate chip or computer.

Data analysis can be performed using standard chemometric

methods such as principal component analysis and SIMCA,

which are available in commercial software packages that

15   run on a PC or which are easily transferred into a computer

running a resident algorithm or onto a signal analysis chip

either integrated onto, or working in conjunction with, the

sensor measurement electronics. The Fisher linear

discriminant is one preferred algorithm for analysis of the

20   data, as described below. In addition, more sophisticated

algorithms and supervised or unsupervised neural network

based learning/training methods can be applied as well

(Duda, R. O.; Hart, P. E. *Pattern Classification and Scene*

Analysis; John Wiley & Sons: New York, 1973, pp 482).

The signals can also be useful in forming a digitally
transmittable representation of an analyte in a fluid.
Such signals could be transmitted over the Internet in
5   encrypted or in publicly available form and analyzed by a
central processing unit at a remote site, and/or archived
for compilation of a data set that could be mined to
determine, for example, changes with respect to historical
mean "normal" values of the breathing air in confined
10   spaces, of human breath profiles, and of a variety of other
long term monitoring situations where detection of analytes
in fluids is an important value-added component of the
data.

15   20-30 different sensors is sufficient for many analyte
classification tasks but larger array sizes can be
implemented as well. Temperature and humidity can be
controlled but because a preferred mode is to record
changes relative to the ambient baseline condition, and
20   because the patterns for a particular type and
concentration of odorant are generally independent of such
baseline conditions, it is not critical to actively control
these variables in some implementations of the technology.

Such control could be achieved either in open-loop or closed-loop configurations.

The sensors and sensor arrays disclosed herein could be
5 used with or without preconcentration of the analyte depending on the power levels and other system constraints demanded by the user. Regardless of the sampling mode, the characteristic patterns (both from amplitude and temporal features, depending on the most robust classification
10 algorithm for the purpose) associated with certain disease states and other volatile analyte signatures can be identified using the sensors disclosed herein. These patterns are then stored in a library, and matched against the signatures emanating from the sample to determine the
15 likelihood of a particular odor falling into the category of concern (disease or nondisease, toxic or nontoxic chemical, good or bad polymer samples, fresh or old fish, fresh or contaminated air etc.).

20 Analyte sampling will occur differently in the various application scenarios. For some applications, direct headspace samples can be collected using either single breath and urine samples in the case of sampling a patient's breath for the purpose of disease or health state

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differentiation and classification. In addition, extended
breath samples, passed over a Tenax, Carbopack, Poropak,
Carbosieve, or other sorbent preconcentrator material, can
be obtained when needed to obtain robust intensity signals.

5   The absorbent material of the fluid concentrator can be,
but is not limited to, a nanoporous material, a microporous
material, a chemically reactive material, a nonporous
material and combinations thereof. In certain instances,
the absorbent material can concentrate the analyte by a

10 factor that exceeds a factor of about 10^5, or by a factor of
about 10^2 to about 10^4. In another embodiment, removal of
background water vapor is conducted in conjunction, such as
concomitantly, with the concentration of the analyte. Once
the analyte is concentrated, it can be desorbed using a

15 variety of techniques, such as heating, purging, stripping,
pressuring or a combination thereof.

Breath samples can be collected through a straw or suitable
tube in a patient's mouth that is connected to the sample

20 chamber (or preconcentrator chamber), with the analyte
outlet available for capture to enable subsequent GC/MS or
other selected laboratory analytical studies of the sample.
In other applications, headspace samples of odorous
specimens can be analyzed and/or carrier gases can be used

to transmit the analyte of concern to the sensors to produce the desired response. In still other cases, the analyte will be in a liquid phase and the liquid phase will be directly exposed to the sensors; in other cases the

5 analyte will undergo some separation initially and in yet other cases only the headspace of the analyte will be exposed to the sensors.

Using the device of the present invention, the analyte can

10 be concentrated from an initial sample volume of about 10 liters and then desorbed into a concentrated volume of about 10 milliliters or less, before being presented to the sensor array.

15 Suitable commercially available adsorbent materials include but are not limited to, Tenax TA, Tenax GR, Carbotrap, Carbopack B and C, Carbotrap C, Carboxen, Carbosieve SIII, Porapak, Spherocarb, and combinations thereof. Preferred adsorbent combinations include, but are not limited to,

20 Tenax GR and Carbopack B; Carbopack B and Carbosieve SIII; and Carbopack C and Carbopack B and Carbosieve SIII or Carboxen 1000. Those skilled in the art will know of other suitable absorbent materials.

65

In some cases, the array will not yield a distinct

signature of each individual analyte in a region, unless

one specific type of analyte dominates the chemical

composition of a sample. Instead, a pattern that is a

5   composite, with certain characteristic temporal features of

the sensor responses that aid in formulating a unique

relationship between the detected analyte contents and the

resulting array response, will be obtained.

10   In a one embodiment of signal processing, the Fisher linear

discriminant searches for the projection vector, w, in the

detector space, which maximizes the pairwise resolution

factor, i.e., rf, for each set of analytes, and reports the

value of rf along this optimal linear discriminant vector.

15   The rf value is an inherent property of the data set and

does not depend on whether principal component space or

original detector space is used to analyze the response

data. This resolution factor is basically a multi-

dimensional analogue to the separation factors used to

20   quantify the resolving power of a column in gas

chromatography, and thus the rf value serves as a

quantitative indication of how distinct two patterns are

from each other, considering both the signals and the

distribution of responses upon exposure to the analytes

that comprise the solvent pair of concern. For example,

assuming a Gaussian distribution relative to the mean value

of the data points that are obtained from the responses of

the array to any given analyte, the probabilities of

5   correctly identifying an analyte as a or b from a single

presentation when a and b are separated with resolution

factors of 1.0, 2.0 or 3.0 are approximately 76%, 92% and

98% respectively.

10   To compute the rf, from standard vector analysis, the mean

response vector, x_a, of an n-sensor array to analyte a is

given as the n-dimensional vector containing the mean auto-

scaled response of each sensors, A_{aj}, to the ath analyte as

components such that

15        $x_a = (A_{a1}, A_{a2}, ... A_{an})$

The average separation, d, between the two analytes, a and

b, in the Euclidean sensor response space is then equal to

the magnitude of the difference between x_a and x_b. The

noise of the sensor responses is also important in

20   quantifying the resolving power of the sensor array. Thus

the standard deviations, $s_{a,d}$ and $s_{b,d}$, obtained from all the

individual array responses to each of a and b along the

vector d, are used to describe the average separation and

ultimately to define the pairwise resolution factor as

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$$rf = d_w / (\ ^2_{a,w}\ +\ ^2_{b,w})\ .$$

Even if the dimensionality of odor space is fairly small, say on the order of 10^1, there is still interest in being

5   able to model the biological olfactory system in its construction of arrays consisting of large numbers of receptor sites. Furthermore, even if a relatively small number (<10) of ideal sensors could indeed span odor space, it is not likely that such ideal sensors could be

10   identified. In practice, correlations between the elements of a sensor array will necessitate a much larger number of sensors to successfully distinguish molecules. Furthermore, performance issues such as response time, signal averaging, or calibration ranges may require multiple sensors based on

15   each material. Analysis of regions will add additional degrees of freedom if the components of the region are to be individually identified and will require large numbers of sensors. Fabrication of large numbers of sensors also enables the use of very powerful coherent signal detection

20   algorithms to pull a known, but small amplitude, signal, out of a noisy background. Because of all of these issues, the number of sensors required to successfully span odor space in a practical device may rapidly multiply from the minimum value defined by the dimensionality of smell space.

The approach described herein uses experimental data (*e.g.* a signal profile, such as a resistance fingerprint) that is generated by an array of differentially responsive sensors.

5   Such sensors include, for example, chemically-sensitive resistor of a sensing array, such as that found in an "electronic nose" as described in USP 5,571,401 (the disclosure of which is incorporated herein), when it is exposed to a molecule of interest. The change in the

10   electrical resistance of a chemically-sensitive resistor in such a sensing array can be related to the sorption of a molecule of interest to, for example, the non-conductive regions of the chemically-sensitive resistor. The signals produced by a plurality of chemically-sensitive resistors

15   having individual sorption criteria thus provide information on a number of chemically important properties, such as the hydrophobicity, molecular size, polarity, and hydrogen-bonding interactions of a molecule of interest, thus, for example, creating a resistance profile or

20   fingerprint of the molecule of interest based upon its chemical properties.

By "molecule of interest" or "analyte" is meant any number of various molecules. For example a molecule or analyte of

interest may be a nucleic acid (*e.g.*, DNA or RNA), a

polypeptide (*e.g.*, an antibody, protein, enzyme), a

biochemical (*e.g.*, a lipid, hormone, fatty acids,

carbohydrate), pharmaceuticals, a chemical such as organics

5 including, for example, alkanes, alkenes, alkynes, dienes,

alicyclic hydrocarbons, arenes, alcohols, ethers, ketones,

aldehydes, cyclic hydrocarbons, carbonyls, carbanions,

polynuclear aromatics and derivatives of such organics,

e.g., halide derivatives.

10

The analysis of a resistance signal pattern (*e.g.* a

resistance profile) of the embodiment may be implemented in

hardware or software, or a combination of both (*e.g.*,

programmable logic arrays or digital signal processors).

15 Unless otherwise specified, the algorithms included as part

of the invention are not inherently related to any

particular computer or other apparatus.

In particular, various general purpose machines may be used

20 with programs written in accordance with the teachings

herein, or it may be more convenient to construct more

specialized apparatus to perform the operations. However,

preferably, the embodiment is implemented in one or more

computer programs executing on programmable systems each

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comprising at least one processor, at least one data

storage system (including volatile and non-volatile memory

and/or storage elements), at least one input device, and at

least one output device. The program code is executed on

5   the processors to perform the functions described herein.

Each such program may be implemented in any desired

computer language (including machine, assembly, high level

procedural, or object oriented programming languages) to

10   communicate with a computer system. In any case, the

language may be a compiled or interpreted language.

Each such computer program is preferably stored on a

storage media or device (*e.g.*, ROM, CD-ROM, or magnetic or

15   optical media) readable by a general or special purpose

programmable computer, for configuring and operating the

computer when the storage media or device is read by the

computer to perform the procedures described herein. The

system may also be considered to be implemented as a

20   computer-readable storage medium, configured with a

computer program, where the storage medium so configured

causes a computer to operate in a specific and predefined

manner to perform the functions described herein.

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EXAMPLE

To test the ability of the "electronic nose" to identify molecules of interest having a particular biological activity selected from a library of molecules of interest, a quantitative structure-activity relationship (QSAR) was used to predict the inhibitory action of a series of alcohols on cytochrome P-450 aniline p-hydroxylation.

Polymer synthesis and preparation. Polymers were generally dissolved in tetrahydofuran, except for poly(4-vinylpyridine) and poly(vinylpyrrolidone), which were dissolved in ethanol, and poly(ethylene-co-vinyl acetate)(18% vinylacetate), 1,2-poly(butadiene), and poly(butadiene)(36% cis and 55% trans 1-4), which was dissolved in toluene. Each polymer (160 mg) was dissolved in its respective solvent (20 ml) either at room temperature or by heating to 35-40 C for several hours. Carbon black (40 mg) was added and the suspension sonicated for at least 20 minutes.

Sensor Fabrication. Corning microscope slides were cut into 10 mm x 25 mm pieces to provide substrate for the sensor. A 7-8 mm gap across the middle of each piece was masked while 300 nm of chromium and then 500 nm of gold was

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evaporated onto the ends of the slides to form the
electrical contacts. Sensors were formed by spin-coating
polymer/carbon black suspensions onto the prepared
substrates. The resulting films were then allowed to dry

5   overnight.

Measurements. An automated flow system consisting of
LabVIEW software, a pentium computer, and electronically
controlled solenoid valves and mass flow controllers were

10   used to produce and deliver selected concentration of
solvent vapors to the detectors. To obtain the desired
analyte concentration, a stream of carrier gas was passed
through a bubbler that had been filled with the solvent of
choice. Saturation of the carrier gas with the solvent

15   vapor was verified through measurement of the rate of mass
loss of the solvent in the bubbler. The vapor-saturated
carrier gas was then diluted with pure carrier gas through
the use of mass flow controllers (MKS Instruments, Inc).
The carrier gas for all experiments was oil-free air,

20   obtained from the general compressed air laboratory source,
containing 1.10 +/- 0.15 parts-per-thousand (ppth) of water
vapor. The air was filtered to remove particulates but
deliberately was not dehumidified or otherwise purified to
reproduce a range of potential "real world" operating

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environments. Calibration of the flow system using a flame ionization detector (model 300 HFID, California Analytical Instruments, Inc.) Indicated that the delivered analyte concentrations were present.

5

Eight bubblers for generation of vapors were available, so the 22 alcohols and 2 diols were divided into 3 groups of 8 as indicated in Figure 5. To pre-condition the sensors, prior to each of the 3 runs, the sensors were subjected to

10 40 exposures, 5 to each of the 8 analytes. Data collection then consisted of a set of 10 exposures to the 8 analytes, with 80 exposures performed in randomized order to eliminate systematic errors from history effects. In the third run, bubbler 2 was replaced by a pyrex tube 37 cm in

15 length with a 1 cm inner diameter. This tube was loaded with approximately 25 cm of granular, solid neopentanol. Flow rates were calculated to give 100 ml/min of saturated vapor from the bubblers, which were of sufficient path length to provide saturated vapors. The background air

20 flow was 1900 ml/min, so that the analyte concentration delivered to the sensors was 5% of the analyte's saturated vapor pressure at room temperature. The ability of the vapor delivery system to provide the expected analyte concentrations based on the input and control settings to

the mass flow controllers as verified using a calibrated

flame ionization detector that sampled several test analyte

gas streams being delivered to the sensor chamber.

5   An exposure had 300 seconds of background air flow,

followed by 300 seconds of flow of analyte at 5% of its

saturated vapor pressure, followed by 300 seconds of the

background air. The DC resistance of each sensor was

measured at intervals of approximately 6 seconds using a

10   multiplexing ohmmeter. The baseline resistance of a sensor

was taken as an average of all measurements of the

resistance of that sensor acquired over a 60 second period

that started between 60 and 66 seconds prior to the start

of the exposure to an analyte. The exact initiation time

15   of this baseline resistance measurement was different for

each sensor, due to small variations in the time interval

required to read the set of 20 resistance values through

the multiplexing ohmmeter. The resistance response for

each sensor to an analyte was taken as an average of all

20   measurements for that sensor in a 60 second period that

started between 234 and 240 seconds after the beginning of

the presentation of the vapor to the sensors, with the

exact initiation time for each sensor channel staggered

similarity to that of the baseline resistance readings. A

response was taken to be the change in resistance of a

sensor, R, divided by its baseline resistance, R. All

differential resistance values (R/R) used in the data

analysis represented, or very closely approximated, the

5   steady-state resistance readings obtained from the sensors

during exposure to the analyte of interest.

Data Analysis. Initial raw data manipulation and

calculation of responses was performed using Microsoft

10   Excel. Multiple Linear regression (MLR) was performed using

either Excel or the QSAR {Define} module of the Cerius2

program (Molecular Simulations, Inc.) on a Silicon Graphics

O2 computer. Many possible MLR models were created,

compared, cross-bred, and evolved by the genetic function

15   approximation on Cerius2.

Results. Figure 2 presents the relative differential

resistance responses for various conducting polymer

composite sensors to three representative alcohols, and

20   Figure 5 summarizes all of the sensor response data for the

various alcohols investigated in this work. Each alcohol

produced a distinct, characteristic response pattern with

the array of sensors chosen for use in the work. Other

sensor arrays comprising different polymer formulatives are

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clearly capable of providing response patterns useful in the present invention.

The responses of the 19 working sensors to 20 of the

5   alcohols (Figure 5) were used to build a QSAR model. Benzyl alcohol and tert-amyl alcohol were excluded from the fit because their biological activities were anomalous. The two diols were also excluded while building the model.

10   The inhibitory action data of Cohen and Mannering (Mol. Pharmacol. 1973, 9, 383-397) are listed in Figure 5. The values are expressed as pI_{50}, where I_{50} is the concentration of the alcohol (in mM) at which the activity of the enzyme is 50% inhibited, and pI_{50} is the negative logarithm of I_{50}.

15   More positive numbers correspond to more strongly inhibiting alcohols.

The QSAR equations consist of a linear combination of descriptors whose coefficients are obtained by a least-

20   squares fitting of predicted to observed biological activity through multiple linear regression. Equation 1 represents a general set of QSAR equations,

$$A \cdot X_{1,1} + B \cdot X_{1,2} + C \cdot X_{1,3} + \cdots + J \cdot X_{1,n} + K = Y_1 \quad (1a)$$

$$A \cdot X_{2,1} + B \cdot X_{2,2} + C \cdot X_{2,3} + \cdots + J \cdot X_{2,n} + K = Y_2 \quad (1b)$$

$$\cdot$$
$$\cdot$$
$$\cdot$$

5 $$A \cdot X_{m,1} + B \cdot X_{m,2} + C \cdot X_{m,3} + \cdots + J \cdot X_{m,n} + K = Y_m \quad (1m)$$

where Y_i is the biological activity of the ith molecule, $X_{i,j}$

is the value of the jth descriptor for the ith molecule, and

A, B, C, . . . K are constants that are obtained through

10   the fitting of Y_i (predicted) versus Y_i(observed). In

Equation 1, the ith alcohol's inhibitory activity is

represented by Y_i and its n sensor responses are taken as

its descriptors ($X_{i,1}$ to $X_{i,n}$).

The genetic function algorithm of the QSAR module of

15   Cerius2 was used to select the best sensors for the QSAR.

One hundred multiple linear regression models were

generated from random combinations of 4 sensors. These

models were ranked according to a lack-of-fit (LOF)

parameter, as given by equation 2:

20
$$LOF = \frac{LSE}{(1-((c+dp)/m))^2} \quad (2)$$

LSE is the least-squares error, c and p are both the number

25   of descriptors (sets of relative differential resistance

response of the sensors in the array) for a simple linear

model such as the one herein, M is the number of samples

(e.g., alcohols), and d is the "smoothing parameter", which

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is entered by the user (1.0 was used). The LOF value is

therefore an inverse measure of how well the model fits the

data, with a penalty for the use of a large number of

descriptors relative to samples. From the set of 100

5   models, two "parents" are chosen, with a probability

inversely proportional to their LOF, and "crossed over"--

some of the descriptors from each are used to form a new

model. There is then a probability for "mutation", where a

new, randomly chosen, descriptor is added to the

10   "daughter". If the daughter is not already present in the

population, it replaces the model with the worst LOF from

the population. After 5,000 rounds of genetic operation,

convergence is generally reached, in which the optimal

models have been found.

15

When the 19 sets of responses from the working sensors were

given to the Genetic Function Algorithm (GFA), a model that

incorporated 5 of the sensors was found to be optimal. The

best fit is described by equation 3:

20

(3) $pI_{50} = 0.51 \cdot 3 + 1.90 \cdot 9 - 3.58 \cdot 13 - 2.14 \cdot 15 - 0.90 \cdot 18 - 1.29$

n=20   R=0.995   s=0.092   F=297

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The numbers in bold refer to sets of responses from the sensors with those numbers, n is the number of samples, R is the correlation coefficient, and s is the standard error. The correlation coefficient of 0.995 indicates that

5 the fit was quite good. The F statistic of 297 indicates that the overall significance of the fit is very high, in fact is at a level of $1-10^{-13}$. Coefficients for all sensors are significant far beyond the 99.9% level, as attested to by their t statistics (see table 3). Predicted versus

10 experimental pI_{50} values are plotted in Figure 3.

TABLE 3

Regression Statistics For the Coefficients of Equation 3

	Coefficient	Standard Error	t Stat	P-value
Intercept	-1.29	0.27	-4.71	3.32E-04
3	0.51	0.07	6.93	6.98E-06
9	1.90	0.19	9.92	1.03E-07
13	-3.58	0.21	-17.13	8.70E-11
15	-2.14	0.27	-7.91	1.56E-06
18	-0.90	0.08	-11.34	1.94E-08

The t statistic is equal to the value of the coefficient divided by its standard error; it is used to derive the P

15 value, which indicates the significance of the coefficient.

Methanol has an inhibition activity distinctly different

from that of the other alcohols, and this can lead to a

misleadingly good fit through a "point and cluster" effect.

A second least-squares fitting of equation 3 was performed

5 with the exclusion of methanol. The coefficient of **15**

changed from -2.14 to -2.20, while those of the other

sensors remained nearly the same. The overall quality of

the fit declined; F decreased from 297 to 109,

corresponding to a decrease in the significance of the fit

10 from the level of $1-(1 \times 10^{-13})$ to $1-(4 \times 10^{-10})$. The

decrease quality of the fit occurs because methanol is

modeled well by the equation, but when methanol is excluded

there is much less variation in the data to be fit.

15 <u>Electronic Nose-Based QSAR</u>. The selection of which

molecules to include in a QSAR is crucial. In the sense,

that it is desirable to use the broadest set of molecules

available to build a QSAR, while not including only one or

two molecules from a distinctly different class of

20 compounds. For example, benzyl alcohol, the only aromatic

alcohol in the data set, has a higher activity than is

predicted by both our QSAR and another QSAR on the

cytochrome P-450 system. The anomalous activity of benzyl

alcohol could be accounted for with an additional

descriptor unique to benzyl alcohol, but the choice of such

a parameter is rather arbitrary, so benzyl alcohol was

excluded during the building of our QSAR. Tert-amyl

alcohol was also excluded because there is evidence that

5 tertiary alcohols function through a stimulatory mechanism

in addition to the usual inhibitory mechanism. As would be

expected in *tert*-amyl alcohol were also acting through this

stimulatory mechanism, its inhibitory activity is

anomalously low. The two diols were also excluded while

10 building the model. Because of these limitations, the QSAR

is expected to be most successful at predicting the

activity of aliphatic mono-alcohols having no other

functionalities.

15 The sensors chosen for the model by the GFA are among those

whose responses are most reproducible. Reproducibility was

measured by examining the set of 10 response of a given

sensor to a given analyte. The value $S_{i,j}$ is defined as the

standard deviation among the 10 responses of the jth sensor

20 to the ith alcohol divided by the average of those

responses. Each sensor has a set of 20 *S* values, one for

each alcohol. A sensor's reproducibility can be gauged by

the median of its set of *S* values. Four of the five

sensors used in the model displayed median *S* values less

82

than 0.063, raking them among the best seven sensors. The only sensor outside this group, **15**, responded only to very polar analytes. Since its response to the majority of the analytes was quite small, its *S* value for those analytes is

5   very large. However, for the analytes to which it did respond, for example methanol and ethanol, its *S* values are small, 0.040 and 0.041, respectively. The inclusion of **15** might be questioned if it were necessary only to model the activity of one analyte, namely the outlier methanol. To

10   test the validity of including **15** in the QSAR, equation 3 was refit with the same set of sensors and all of the previously used alcohols, excluding methanol. In the new QSAR, the significance of **15** remains significant. If the set of five sensor responses to methanol are substituted

15   into the second QSAR equation, which was formed with no information about methanol, the predicted pI_{50} of methanol is -3.12 very close to its experimental value of -3.09. It appears that whatever molecular characteristics are probed by **15** are successfully extrapolated from the more

20   moderately polar analytes to methanol. In other words, **15** is not just an indicator variable for methanol that is fit with an arbitrary coefficient.

A quantitative measure of the predictive power of the QSAR

can be obtained by building a model using the biological

and sensor response data from all the molecules except one,

and then predicting the activity of the excluded molecule

5   with that model. The procedure is repeated for each

molecule in the data set, and the predictive sum of squares

(PRESS) is defined as the sum, over all analytes, of the

squared differences between the predicted and actual

biological activity. Using equation 3, the PRESS for the

10 set of 20 alcohols is 0.221. This value can be compared to

the residual sum of squares, RSS, in which one QSAR

equation (fit to all samples) is used to calculate the

predicted activity. As would be expected, the RSS of 0.117

is lower than the PRESS. More significantly, a large

15 difference between the PRESS and RSS would imply that the

model had used too many parameters and overfit the data,

and this appears not to be the case.

An optimum fit (as judged by the LOF parameter) was found

20 to require five descriptors; no equation with a different

number of descriptors formed as significant a model. The

best 4 sensors QSAR, consisting of sensors **1**, **13**, **16** and

17, has an R=0.984, s=0.163, and F=114, indicating an

overall significance at the level of $1 - (5 \times 10^{-11})$. On

84

the other hand, addition of further sensors adds parameters and enables a better fit to the data set. However, if **4** is added to equation 3 to form the best 6-sensor equation, certain key statistics point to a diminished model. As

5   would be expected with an additional parameter, R increases, from 0.995 to 0.996. Additionally, the standard error decreases from 0.0916 to 0.0834, the RSS decreases from 0.117 to 0.090, and the F statistic increases from 297 to 300. However, the significance of the fit, represented

10   by the F statistic, decreases from $1 - (1.08 \times 10^{-13})$ to $1 - (3.66 \times 10^{-13})$. The PRESS increases from 0.221 to 0.253. Thus, although the 6-sensor model fits the set of 20 alcohols better than the 5-sensor model, the 6-sensor model is worse at predicting the activity of an alcohol that was

15   not included in the fit, indicating that the 6-sensor model has overfit the data.

As described above, the cytochrome P-450 p-hydroxylation inhibition activities of all the aliphatic mono-alcohols

20   investigated in this work could be quite accurately predicted from a model that was constructed without the use of any information about the molecular structure of the alcohols for which the prediction are made. This indicates that the resistance data output of the electronic nose

contains implicit information on most of the chemical factors that control the interactions of the enzyme with the alcohols. These resistance data reflect the binding interactions between the alcohols and a collection of

5 polymers having a diverse collection of chemical attributes. It is not necessary that an individual polymer probe specifically and exclusively one such descriptor of the analyte-substrate interaction, because the desired information can be obtained through analysis of the

10 collective response of the sensor array to an analyte.

Comparison with Other QSARs. Cohen and Mannering fit the activity of 11 of the unbranched 1- and 2-alcohols (excluding methanol) to a one parameter equation using log

15 P (J. Mol. Pharmaco. 1973, 9, 383-397). A modified version, using updated log P values and fit to only 10 alcohols (excluding methanol and ethanol), was given later by Shusterman (equation 4) (Chem.-Biol. Interactions 1990, 74, 63-77).

20

$$pI50 = 0.43 \log P - 0.53 \qquad (4)$$
$$N=10 \qquad R=0.954 \qquad s=0.128$$

However, Shusterman also showed that for a larger set of

25 alcohols, a simple fit to log P was inadequate to describe

most of their activity; a fit of 19 alcohols yielded

equation 5, which has rather poor regression statistics.

$$pI50=0.35\log P-0.71 \qquad (5)$$
$$n=19 \qquad R=0.505 \qquad s=0.468$$

5

In a second equation using two descriptors, log P and (log

p)^2, Cohen and Mannering fit 17 of the alcohols with an R

of 0.98 (equation 6).

10

$$pI_{50}=1.50 \ \log P \ -0.36(\log P)^2+1.75$$
$$n=17 \qquad R=0.98 \qquad s=0.44$$

Although this was a better fit, it used more descriptors.

15 Additionally, it is evident from inspection of the data

that there are factors besides hydrophobicity that

determine an alcohol's activity. Four subsequent QSARs

have therefore been used to model; the data set more fully

and some aspects of these models are discussed below.

20

A more complex, three parameter, QSAR was based upon logP,

a calculated electronic parameter ($_{HOMO}$), and a steric

parameter (BULK$_{1at}$)(equation 7).

25 $$pI_{50}=16.2\log P-16.0\log(\ P+1)-1.35BULK_{1at}+0.381 \ _{HOMO}+22.5 \qquad (7)$$
$$n=21 \qquad R=0.982 \qquad s=0.170 \qquad \log =1.05$$

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Shusterman and Johnson, however, pointed out that the use

of $_{HOMO}$ as a parameter was unjustified since it was

necessary only to fit benzyl alcohol, and becomes an

insignificant parameter (as indicated by its t value) when

5   benzyl alcohol is excluded from the data set. Similarly,

the bilinear dependence of pI50 upon log P of equation 7

was necessary only to fit a single data point, methanol.

Another QSAR, based on a choice of molecular connectivity

10   indices, has also been used to model the activity of 20

alcohols (benzyl alcohol and tert-amyl alcohol were

excluded (equation 8).

$$pI50 = -6.88(1/^0\chi^v) - 1.14\ ^4\chi_{PC} + 1.85 \tag{8}$$
15   $n=20 \qquad R=0.983 \qquad s=0.156$

The parameter $^0\chi^v$, the zero-order valence molecular

connectivity index, basically corresponds to molecular

size, and therefore hydrophobicity, for this set of

20   molecules. Hence, the inverse of the index has a negative

coefficient in equation 8. The parameter $^4\chi_{PC}$, the fourth-

order path/cluster molecular connectivity index, correlates

with the degree of branching in the molecule, and therefore

also has a negative coefficient in equation 8.

25

A third QSAR, which relies entirely upon calculated

electronic parameters as descriptors, has been constructed

and used to fit all 22 alcohols. Shusterman noted problems

with the QSAR. For example, it was asserted that the -

5   carbon of the alcohols was acting as an electron acceptor

from the enzyme, because a correlation between activity and

QCL, the electron density on the -carbon in the LUMO, was

found. QCL is correlated with log P(R=0.747), to some

extent explaining the fit. Two alcohols, 3- methylbutanol

10   and 2,4-dimethyl-3-pentanol, were poorly fit, and no

rationalization was presented for why the correlation with

QCL would not apply to these two substrates as well.

Finally, Shusterman created a QSAR based on log P and two

15   steric parameters, M and A, which were used to describe the

branching of the alcohols. M is the number of carbons

beyond the methyl substituent in Figure 3, thus, 1- and 2-

alcohols have an M=0, while M for 3-pentanol would be one,

and for 2,4-dimethyl-3-pentanol is 2. The second

20   parameter, A, refers to the number of branched carbons in

the main chain, A=1 for 2-methyl-1-butanol and 2 for

neopenyl alcohol. A fit of 19 of the alcohols (benzyl

alcohol, tert-amyl alcohol, and methanol were excluded)

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yielded equation 9. The negative coefficient for M and A

indicate the loss of activity with branching.

$$pI50=0.48logP-0.65 \cdot M-0.31 \cdot A-0.60 \qquad (9)$$

5   n=19   R=0.955   s=0.171

To compare the electronic nose QSAR to those of Sabljic and

Shusterman, one must use statistics that take into account

the number of descriptors used. Table 5 lists the

10   comparison of selected regression statistics from the QSAR

of Sabljic, Shusterman, equation 3, and the QSAR created

when the coefficients of equation 3 were fit to the 19

alcohols besides methanol (R is the correlation

coefficient, s is the standard error, and the final column

15   is the overall significance of the regression equation).

Because the electronic nose QSAR model uses more

parameters, it is inappropriate to compare just either the

correlation coefficients, standard error, or residual sum

of squares of the models. To some extent, the PRESS should

20   be independent of the number of parameters in a model,

since the model is tested upon molecules about which it has

no information. The PRESS of the electronic nose QSAR

model is significantly lower than the other two models of

interest. Finally, the F steatitic gauges the overall

25   significance of the fit while accounting for the number of

90

parameters used. By this measure, the electronic nose QSAR

is approximately as significant as Sabljic's and more

significant than Shusterman's.

Table 5:

	Data pts fit	descriptors used	R	s	RSS	PRESS	F	Significance F
Sabljic	20	2	0.983	0.156	0.414	0.872	250	2.51E-13
Shusterman	19	3	0.956	0.17	0.436	0.786	53	3.34E-08
Present Disclosure	20	5	0.995	0.092	0.117	0.221	297	1.08E-13
Present Disclosure (no methanol)	19	5	0.988	0.095	0.117	0.243	109	3.89E-10

5

It appears that the important chemical interaction involved

in the partitioning of the aliphatic alcohols into the

10   enzyme binding site are probed by the array responses. The

construction of our QSAR did not require making assumption

regarding which steric or electronic factors are important

or what parameters to use to capture such effects.

Obtaining chemical insight into the nature of the dominant

15   binding forces involved in the reaction being modeled would

require a complete understanding of the chemical factors

that determine the analyte partitioning into each polymer

in the electronic nose. In principle it is possible to

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extract such information for certain descriptors of

interest, but it is not necessary to have such information

in order to use the readily-obtained electronic nose data

to predict successfully the activity of various alcohols in

5   inhibiting cytochrome P-450 activity.

MATERIALS

This example illustrates the use of an array of

conductive/non-conductive region sensors. In this example,

10 carbon black is the conductive region material. The carbon

black (Black Pearls 2000) is a furnace black material from

Cabot Co. (Billerica, MA). The nonconductive region

comprises a nonconductive polymer.

15 To prepare the detector substrates, two parallel bands of

gold, 50-100 nm thick and separated by 5 mm, are deposited

onto conventional 7.5 cm x 2.5 cm glass slides. The slides

are then cut into strips to produce 0.7 cm x 2.5 cm pieces

of glass, with each strip of glass having one pair of Au

20 leads spaced 5 mm apart.

The detectors are made from a solution of polymer into

which carbon black has been suspended. In this example,

125 mg of the polymer is dissolved in 10 mL of

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tetrahydrofuran, and carbon black (42 mg) is then suspended in this solution, to produce a composition of 75% polymer and 25% carbon black by weight of solids.

5   One coating of this suspension is applied to each substrate yielding a film thickness of ≈1 micron as can be determined by atomic force microscopy. For larger sensor thicknesses, more coats are used.

10   METHODS

The dc resistance of each detector is determined as a function of time using a simple two-point resistance configuration. Contacts are made to the gold lines by pressure-contacting electrical leads using flat-jawed

15   alligator clips. Resistance data is acquired using a Hydra 2620A Data Acquisition Unit (John Fluke Mfg. Co.; Everett, WA), which is interfaced to a personal computer. All of the films had resistance values below the 10 MΩ limit of the Hydra 2620A.

20

To initiate an experiment, five copies of a given detector type are placed into the glass chamber and a background flow of nitrogen is introduced until the resistance of the detectors is stabilized. Solvent vapor streams are then

passed over the detectors. The background and analyte flow

rates are monitored using two flow meters (Gilmont

Instruments, Inc.) which have limits of 0.2 L min^{-1} to 15.0

L min^{-1} and 0.0015 L min^{-1} to 0.310 L min^{-1}, respectively.

5   In a typical experiment, resistance data on the detectors

are collected for 150 s with just the background gas

flowing (typically about 1 - 2 L min^{-1}) to serve as a

baseline. This is followed by a 10 s to 2 minutes data

collection while the detectors are exposed to the analyte

10 vapor stream (typically about 200 - 300 mL min^{-1}). The

detectors are then given 200 - 300 s to recover during

which pure background gas was passed through the chamber.

The exposure times varied, but steady state values of

resistance change are always reached for any given exposure

15 time. Resistances for all detectors in a given trial were

monitored contemporaneously through the use of the

multiplexing capabilities of the Hydra voltmeter. Results

are obtained by running two trials of five exposures each,

with the trials performed on different days. Each analyte

20 is exposed to five copies of the detector simultaneously

and the results.

This example illustrates a response of an array of sensors having predetermined polymer thicknesses of 100 nm and 500 nm to an analyte.

5   With reference to Figure 6, a response of an array of detectors having predetermined polymer thicknesses of 100 nm and 500 nm to an analyte is illustrated. In this example, the polymer used was polycaprolactone and the analyte used was benzophenone. All the detectors displayed

10   an increase in resistance upon exposure to the vapor, and returned to their baseline values after the vapor was removed. The change in resistance for the thinnest polymer showed steady-state conditions faster compared to the thickest sensor which reach steady-state conditions at a

15   slower pace. The responses are analyzed by calculating the maximum differential response value, ΔR_{max}, observed during the exposure period and dividing it by the baseline value of the resistance, R_i, (taken as the resistance value just before the exposure began) and expressed as:

20                                    $$\Delta R_{max}/R_i$$

                                                                Equation III

A plot of the response flux as a function of time can be used to calculate a diffusion coefficient. In order to measure the diffusivity experimentally in a polymer based

sensor, the time lag procedure is used. The time lag θ is estimated from the time axis intercept and the diffusion coefficient is obtained using Equation II.

This example illustrates a response of an array of

5   detectors having predetermined polymer thicknesses and analytes having different volatilities.

With reference to Figure 7, the responses of an array of detectors having predetermined polymer thicknesses (one

10   thin, one thick) and to different analytes is illustrated. The polymer used was polyethylene oxide and the analytes were hexane and decane. The results indicate that the thin sensor shows steady-state conditions faster compared to the thick sensor which reaches steady state conditions at a

15   slower pace. In addition, the analyte that is more volatile, (hexane), reaches steady-state conditions faster than an analyte that is less volatile (decane). Table 1 shows the responses of the thin and thick sensors versus time as well as the responses of two different analytes.

20

TABLE 5

Time (min)	1	2	4	6	8
Hexane (thin)	0.008917	0.008389	0.007985	0.007678	0.007468
Hexane (thick)	0.004428	0.0062	0.008713	0.010352	0.011586
Decane (thin)	0.007624	0.009453	0.010664	0.011088	0.011286
Decane (thick)	0.001577	0.002578	0.004103	0.005290	0.006279

By monitoring the temporal response lag between different film thicknesses to a response, sensors having advantageous features are realized. For example, when monitoring certain volitilities components, the lag time is an important component which can be monitored. False rates can be minimized if the lag profile does not correspond to the expected lag profile.

Although only a few embodiments have been described in detail above, those having ordinary skill in the art will certainly understand that many modifications are possible

in the preferred embodiment without departing from the

teachings thereof. All such modifications are intended to

be encompassed within the following claims.